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SUPPLEMENTUM IX

EXPERIMENTAL STUDIES ON ROENTGEN TREATMENT OF MALIGNANT TUMORS

BY

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PREFACE.

The investigations reported in this paper[1] were carried out in 926—27, while I was Assistant Physician to the *Roentgen Clinic f Aarhus Kommunehospital* and associated *Radium Station*.

The work was taken up on suggestion from my chief of that ime, Dr. med. CARL KREBS, who by his own investigations had aid the foundation upon which I could plan and carry out my nvestigations. It was kept up under his constant encouragement, nd my completing the work is largely due to the extraordinarily ood conditions for experimental research which he and the li- eral and broad-minded municipal authorities have created in- Aarhus. The city of Aarhus has most generously paid all the xpenses connected with the experiments.

In the course of my experiments the whole personnel of the Roentgen Clinic has been of the greatest help to me by assisting ne in the Roentgen treatments, that often take a great deal of ime, and in many other ways.

To every one who in some way or other has lend me a hand in arrying through this work, I give my hearty thanks.

J. J.

This paper was submitted to the Faculty of Medicine, Uni- ersity of Copenhagen, in June, 1928.

[1] Translated from Danish by HANS ANDERSEN, M. D., Copenhagen.

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I. INTRODUCTION.

Whether the successful outcome of radiotherapy in malignant tumors is contingent upon the direct injury to the tumor cells, or whether it is more dependent upon the tissue reaction elicited by radiation in the surroundings of tumor, that has been a burning question in recent years.

Several investigators have shown that the cell destruction of animal tumors *in vitro* and *in situ* — incapacitating them for growth after implantation in other animals — requires a dose considerably larger than may generally be safely given in the therapy. In addition it was found that irradiation of an area where transplantation of a non-irradiated tumor was later performed, would prevent or inhibit the growth of tumor, and that tumor in such experiments undergoes the same histological changes as are observed after direct radiation. From the view-point that the effect of radiation is indirect to a large extent, it was not improbable that, in the therapy, very large doses would injure the surroundings of tumor to such a degree that the therapeutic result would be less favorable than that obtained with smaller doses. The findings in post-operative irradiation of cancer of the breast, where the large massive doses have given less favorable results than have the more moderate doses, might be interpreted in this way.

My former chief, Dr. CARL KREBS, called my attention to some investigations by KOK & VORLAENDER (Freiburg in Br.), reported in »Strahlentherapie», 1923—24, in which the authors found these particulars confirmed on irradiation of transplantable tumors in mouse. On local radiation of tumor they obtained the best effect with doses approximating the epilation dose for mouse, and they found the effect decreasing with increase of dosage. They further found that radiation of a large field around the tumor gave better results than did radiation of a small area; and they obtained the best effect by exposing the whole animal to a considerably smaller dose.

I purposed thoroughly to investigate these findings on the excellent material furnished me by my chief at the X-ray Clinic, Muni-

cipal Hospital, Aarhus. After many efforts C. KREBS had at that time succeeded in obtaining two different transplantable tumors which appeared to be very suitable for the contemplated experiments.

The problems I decided to investigate were:

1. What are the therapeutic effects of local irradiation with different doses?

2. What is the effect of universal irradiation of the animal?

Having investigated these problems I went on to investigate the difference in effect of the same dose when given as one massive dose and given in several portions. Until now it has been a much disputed question in the clinic whether it is more expedient to treat malignant tumors with massive doses, or with protracted radiation and this question can by no means be regarded as settled.

Like the clinical experiences, the experimental results are conflicting with reference to the influence of the time factor. Some authors have obtained the greatest effect with massive doses, others with protracted radiation. But few investigations of this particular question have been carried out on malignant tumors in animals, and their results do not allow of any definite conclusions.

Therefore, the third problem I proposed to tackle was:

3. What is the difference in effect of the same X-ray dose when given as a single massive dose and when given in several smaller doses at varying intervals?

In the course of this study it was found that fractional radiation showed considerably slighter effect on the skin of the animal than did the single massive dose; the former did not cause epilation while the same massive dose nearly always made the hairs come off. To be sure, the effect of the fractional radiation method on tumor was found to be less than that of the single massive dose; but on the other hand, there was the possibility that this loss in effect might be more than counterbalanced by the absence of injury to the skin, so that the best therapeutic effect really would be obtained with the fractional doses. The question was:

4. When radiation is given in such doses — partly as a single massive dose, partly as several smaller doses — that the skin effect of either treatment is the same, which method will then give the best effect on tumor?

Concurrent with my investigation of this problem I made a series of experiments that were inspired by the paper read by PFAHLER before the First International Congress on Radiology, London 1925. In this address PFAHLER emphasized the excellent clinical results from a radiation method that was first indicated by KINGERY

and by him was called the »saturation method». Later I shall discuss this method in detail; here it suffices to mention that the principle of the method is to give a rather large dose at once — in one or several exposures — and additional smaller doses, so that the tissue reaction is maintained at the same height for some time — without ever getting so marked as to injure the normal tissues.

Then my last problem was this:

5. *When the skin effects are kept alike, will the saturation method give a stronger effect on tumor than do the other methods?*

II. ON RADIOTHERAPY IN MALIGNANT TUMORS IN MAN — WITH SPECIAL REFERENCE TO THE QUESTIONS FORMULATED IN THE INTRODUCTION.

1. Brief Historical Survey of the Radiation Methods.

In the early days of Roentgen therapy only superficial tumors were treated — usually with small daily, or frequently repeated, doses until the desired result was obtained, or until the skin reaction became so severe that it was necessary to discontinue the treatment.

After the introduction of the first practicable measuring methods there was a tendency to make the radiations more concentrated. X-rays were given in larger doses and at longer intervals. The severe injuries to the skin, that were so frequent in the early days, were now less frequent; and the treatment of superficial carcinomata gave often remarkably good results, some of which can stand comparison with the results of our days (NOIRÉ, 125).

These encouraging results soon brought about that also the treatment of deeper located tumors was attempted. In the following years the aim of the efforts was principally to find out how to obtain the greatest depth effect. Radiation was given through several portals of entry, X-ray filters were introduced, the distance from focus to patient was increased, and the field of exposure was enlarged, while the physicists kept on improving the machines and tubes so it became possible to give a constantly harder ray quality, and to give it within a short time. In this work the German clinics in Freiburg and Erlangen were the leading, and it was particularly the Germans, too, who laid more and more stress upon the importance of giving the maximal dose within a minimum of time. From Germany this view gained ground throughout the world, although the French were somewhat more reserved and stood out for the more protracted radiation. The reaction toward this view, however, was not slow in getting started, and it gathered way, particularly after PERTHES had urged to compile the results from postoperative X-ray treatment of cancer of the breast. In this way

it was found that massive radiation with large doses of hard rays gave less favorable results than did the more cautious dosage. The intensive treatment did also loose ground on other points. A few are still adherents of this method, but most radiologists have now gone back to more protracted radiation methods.

While the intensive method is chiefly a German product, the most recent ways have been indicated chiefly by the French — especially through the works by REGAUD. Efforts are made to utilise the assumed special sensitivity of tumor cells during the process of division, in the radium therapy through radiation of low intensity, in the Roentgen therapy be means of frequent small doses. The methods of various radiologists differ a great deal as to minute details of technique, but common to all of them is their tendency to prolong the period of treatment.

A special form of protracted radiation is the saturation method, advanced by KINGERY and introduced in the deep-therapy by PFAHLER. Its principle has been mentioned, and the method will be further discussed later on. It is a combination of the intensive method and the protracted radiation.

These methods bring us up to the present day, when one of the most disputed problems of the radiotherapy is the question of massive or protracted treatment.

2. On Size of Dose in Single Exposure and Massive Radiation.

The term »stimulating dose» plays an important rôle in the earlier works on radiotherapy. It was the general opinion that small doses were capable of stimulating the growth of tumor. In later years there is not much fear of stimulating the cancer cells to more rapid growth by giving small doses; many authors deny the existence of such a stimulation. At any rate, it cannot be regarded as proved. SEITZ (161) estimates the stimulating dose to be 35—40 % HED. Most other authors, however, who do not deny its existence, take it to be considerably smaller. I shall not here enter into the particulars of the numerous papers on the stimulating effect of Roentgen and radium rays but refer to the large work by CZEPA (30), 1924.

The upper margin of dosage is a natural limit in so far as the dose must not exceed what can be sustained by the normal tissues without any greater damage. But — is the maximal dose preferable, or do the middle-size doses give just as good or even better results?

After 1916, when FRIEDRICH & KRÖNIG (42) formulated their idea of the »carcinoma dose», the question of the most suitable dose became very actual. The authors define carcinoma dose as the dose »die eben das Karzinom deutlich zur Rückbildung bringt». In a subsequent paper (82), 1918, they give a slightly different definition: »Die Dosis, welche in einer einzigen Sitzung appliziert, einen Krebs für das Auge und das Gefühl verschwinden macht.» They measured the dose with the ionto-quantimeter; and in cancer of the breast they found it to be 80 % HED. They have also made cancer of the cervix disappear with this dose. They state, however, that there is some difference in sensitivity.

Later SEITZ & WINTZ (162) determine the carcinoma dose by measuring it in cancer of uterus and find it to be 100—110 % HED. As there is some difference between Freiburg HED and Erlangen HED, the two estimates really agree fairly well. SEITZ & WINTZ define the carcinoma dose as the dose it takes to destroy the cancer cells. There may be variations, they think, but not to any great extent. In their later reports they repeatedly call attention to the fact, that their carcinoma dose is not the same as the curative dose, since the latter depends on the condition of the patient, on whether the cachexia is far advanced or not. As late as 1927, WINTZ (181) claims that this dose is sufficient in cancer of uterus and cancer of the breast and also in most other carcinomata. The author found no instance in which it took more than 115 % HED to destroy the cancer cells. — A sarcoma dose has been estimated in similar way; it is said to be somewhat smaller than the carcinoma dose.

This view of a destructive dose common to all carcinomata was met with many objections; now it is discarded by most radiologists. On the other hand, most authors are agreed that it is necessary to give the mentioned dose before tumor may be expected to disappear. Carcinoma dose has now become the minimum required for this effect (EWING, 35, HALBERSTAEDTER, 48, HOLFELDER, 56, JÜNGLING, 66, LEDOUX-LEBARD, 93, MIESCHER, 104, OPITZ, 126, PERTHES, 135).

It then became the general opinion, particularly in Germany, that all cancer tissue ought to be penetrated homogeneously with as large a dose as could be given without serious injury to the surrounding normal tissue. The literature may perhaps contain a few reports of carcinomata that have disappeared on doses less than the HED; but they are exceptions. Most authors of that period are agreed that the best results in the treatment of cancer have been obtained after the introduction of larger doses (BUMM & WARNEKROS, 20, GAUSS, 43, HOLFELDER, 56, JÜNGLING, 68, MIESCHER, 104, PERTHES, 133, 134, WARNEKROS, 174, WETTERER, 179).

In 1920, OPITZ even considers 160 e being too little, while, on the other hand, he regards 250 e as being too much (170 e = 1 HED). Even at that time, however, a few radiologists were claiming that the middle-size doses gave the best clinical results (THEILHABER, 169).

Yet, it was not until after the publication of the results of post-operative radiation in cancer of the breast in the Tübinger Clinic (PERTHES, 135, NEHER, 124) that it was fully realised that large massive doses may also have an aggravating effect. Subsequent reports from numerous clinics (cf. PERTHES, 134) have further established the fact that these doses merely give a higher percentage of recurrence, while it was found that more frequently repeated smaller doses most likely would diminish the number of recurrencies (ANSCHÜTZ & HELLMANN, 7, PERTHES, 134). In the case of Scandinavia these facts have been brought out very clearly by LARSÉN & LYSHOLM (90) in their account of FORSSELL's material.

For explanation of the poor results from the large massive doses the view came into existence that these doses injured the whole organism, or the normal tissues surrounding the cancer, so as to break down the natural defense of the organism against the cancer; and many radiologists began to give up the intensive therapy, as they either began to distribute the dose over a longer period, or gave a smaller total dose in each series. This change in dosage took place not only in the post-operative treatment of cancer of the breast but also in other conditions. OPITZ (127, 128), in particular, has constantly asserted that the large doses also gave less favorable results in the treatment of fully developed tumors on account of injury to the surrounding connective tissue, whose reaction he considers the most important factor in the healing process. This view he found to be confirmed on after-examination of patients who had previously been treated with large doses in the Freiburg Clinic. Nearly all of these patients showed recurrency.

THEILHABER (169) had somewhat similar experiences. In the treatment of cancers of the skin HAZEN & WHITMORE (51) found that »1½ unit doses are as effectual as 2½ unit doses». Nor does EWING (36) think that the results will be better on increase of dose. In radium treatment of cancer of uterus and other carcinomata LABORDE (85) thinks she has seen the best effect from middle-size doses and less effect from very large doses.

Contrary to these authors are others who are still of the opinion that the best results are obtained with large doses — at any rate apart from the prophylactic after-treatments.

HALBERSTAEDTER (48) had a patient who had been treated by somebody else for cancer of the breast. She presented some very

distinct areas in which the skin showed marked injuries, even Roentgen erosions. No tumor nodules, however, could be made out in these areas, whereas such nodules were numerous in a narrow stripe between the areas; and yet, this stripe must have been exposed to a non-inconsiderable dose through scattering. He has seen several cases of that kind.

JÜNGLING (68) has treated a patient suffering from skin metastases after cancer of the breast with radiation of various areas with different doses, ranging up to 120 % HED. The nodules disappeared after 120 % HED, diminished on 100 %, and were not affected by smaller doses. On subsequent irradiation of the whole surface with 140 % nearly all of the nodules went away. Only a 3 mm. wide zone that was covered with lead, and in which the radiation through scattering must have been 60 % at the least, did not show any effect. On repetition the experiment gave the same result.

DEHLER (31) has also observed a rapid growth of cancer metastases in a narrow zone between two fields; and one side of the field, where the distance had been a little larger, showed less effect than did the rest.

PERTHES (134) has seen no effect from treating a series of skin cancers and cancers of the breast with small doses.

MIESCHER (104) treated a number of rodent ulcers with different doses and obtained the following results:

Dose in S.-N.	Number of cases	Permanent cure after 1 applic.
1	3	0
2	4	0
3	2	0
4	9	2
5	7	2
6	7	5
7	13	12

3 S.-N. are equal to 1 HED.; 2—4 mm. Al. filter was used.

SCHMITZ (157) has investigated this question in 418 cases of cancer of the uterus that were treated with radium and X-rays in fractional dosage. He found that the percentage of cure increases with increase of dose, and that the various histological types of tumors differ in sensitivity. The following table gives the percentages of primary healing after the different doses:

	80 % HED.	100 % HED.	130 % HED.	150 % HED.
Spinocellular C.	14 %	25 %	66 %	80 %
Basocellular C.	—	50 %	—	—
Adenocarcinoma	—	0	66 %	—

In 1928, Belot (16) states, with reference to the treatment of cutaneous epitheliomata, that the results are bettered according as the doses are increased. The most favorable statistics were reported by those who gave the largest doses. The upper limit lies within the doses that cause irreparable changes in the normal tissues.

Thus the clinical results seem to indicate that in pronounced tumors the effect of radiation given within a short time increases as the dose is increased to something above 1 HED, where such a dose can be given without injury to important organs or without permeating such large parts of the organism that its resistance is lowered. As to the prophylactic after-treatment in cancer of the breast, it seems most reasonable, in view of the preceding, to assume that the increased recurrence percentage after the large massive doses is not derived from any local injury to the connective tissue, but that it is due to a universal lowering of the resistance of the organism.

3. *On the Significance of the General Effect of Roentgen Rays.*

It has long been known that the action of Roentgen rays is not limited to the site of application, and that they also affect the organism as a whole. The indisposition that is so frequent after X-ray treatment — with nausea, vomiting, and the like, the German »Röntgenkater» — is usually attributed to such a general effect. There are numerous reports on changes in the blood and the blood corpuscles after X-ray treatment.

It is of particular importance to this investigation whether there be any record of malignant tumors that have been cured, or made disappear, with irradiation of any field other than tumor or with universal irradiation.

I have not been able to find one single case in which a malignant tumor was cured on such conditions. Holfelder (57) and Perthes (134) also deny the occurrence of such cures. Isolated cases have been reported in which there was a decrease in size of the metastases after the primary tumor had disappeared on irradiation. But, such findings are also recorded after resection of the primary tumor. They are of no consequence when compared with the large number of cases in which the reverse takes place.

After reviewing the literature Voorhoeve (172) arrived at the conclusion that no definite far-effect of X-rays has been observed. It is a matter in dispute whether such far-effect does really exist or not. He reports himself 3 cases of lymphatic leukemia and 2 cases of malignant granuloma in which, he thinks, he has observed a

definite decrease in size of non-irradiated lymph-glands concurren with a diminution of the irradiated glands. In the systemic dis eases of the lymphatic system, however, one is dealing with condi tions differing from malignant tumors.

KUHLMANN (83) has summed up the literature on this subjec in 1925. He, too, arrives at the conclusion that there has been ob served no definitely favorable result from the far-effect on malignan tumors.

I shall not enter into the far-effect of radiation on endocrine or gans. Here, too, the findings are very contradictory.

A. v. LIEBENSTEIN (95) has reported the results from experi ments with universal radiation on human beings, a few isolate instances of malignant tumors and lymphogranulomatosis. He think he has seen some metastases and glands decrease in size on thi treatment, but he found no favorable effect in most of the cases Nor does he think he has seen any harmful effect of the treatment yet, one of the patient died from a purpura-like disease.

So there is no proof of any favorable influence from the genera effect of irradiation, whereas several things go to show that it ma have an injurious influence.

The findings in after-radiation of cancer of the breast are ex plained in this way by most of the authors (PERTHES, 134, BÉCLÈRE 14, HOLFELDER, 59): The large doses of hard rays on the larg fields permeate such large parts of the organism that it looses som of its resistance toward the cancer. The direct local injury to th connective tissue, however, is also considered contributory to th poor result.

FRIEDRICH & BEHNE (41) make a comparison between cases o cancer of the uterus that were treated with vaginal irradiation an cases treated through ordinary abdominal and dorsal exposures and they arrive at the conclusion that radiation through large parts of the body has an unfavorable influence on the result *i loco*, and more so the larger the permeated volume.

B. DEN HOED (54) reports from Niederländisches Institut fü Krebsforschung that the treatment of cancer of the uterus gav less favorable results when radiation on many small fields was re placed by irradiation after Dessauer's method — with 4 large field whereby most of the abdomen was irradiated.

Perusing the Roentgen literature of later years one finds thes points emphasized again and again. Thus BÉCLÈRE (14) defines th aim of Roentgen therapy as: Destruction of the tumor cells, integrit of the sound surroundings, maintainance of the resistance of th organism. PERTHES (134) speaks of the dual aim of Roentge

therapy: The greatest possible injury to the cancer cells and the greatest possible care of the defensive forces of the organism.

The clinical experiences then leave no basis for assuming a favorable effect from universal irradiation in malignant tumors; many findings, on the other hand, indicate that universal radiation, or radiation of a large part of the organism, may have an injurious effect.

4. On Irradiation with Fractional Doses.

The frequent small doses given in the earliest days of the Roentgen therapy were soon replaced by most radiologists with larger doses at longer intervals. After KRÖNIG & FRIEDRICH (82) had demonstrated that there is a considerable loss in effect on the human skin when the dose is distributed over a longer period, it was made an absolute requirement — particularly in Germany — to treat malignant tumors with the maximum of dose in the minimum of time. And, besides, the fear of stimulating the cancer cells to more rapid growth with the small doses and of rendering them resistant to the radiation effect was also conducive to discontinue the frequent small doses.

Reports on the results from post-operative irradiation in cancer of the breast, however, showed that the intensive treatment is not always the best therapy. For the intensive treatment was found to increase the recurrence percentage, while the smaller and more frequent doses would lower the percentage of recurrence (PERTHES, 134, JÜNGLING, 69, ANSCHÜTZ & HELLMANN, 7, LARSÉN & LYSHOLM, 90, BÉCLÈRE, 15).

Beside the interpretation of these results as due to the beforementioned injurious influence of the large doses on the organism, JÜNGLING (69) also found their explanation in his experiments with irradiation of dry and sprouting beans. In the dry beans he found a complete cumulation of several smaller doses, and an incomplete cumulation in the sprouting beans. After operation, he thought, the latent cancer germs would then behave like the dry seeds, while the progressive tumors would react like the sprouting seeds. And for this reason he still considers the intensive treatment to be the most effective in advanced tumors. BÉCLÈRE (15) and HOLTHUSEN (62) subscribe to this view. PERTHES (134) thinks that just like the large doses lower the resistance of the organism to the invasion of the cancer, so the repeated smaller doses might possibly increase this resistance — in analogy with the conditions that have been demonstrated through animal experiments, particularly by the

English investigators. I shall come back to these experimen later on.

In recent years the use of intensive irradiation has steadily bee loosing ground, not only in the prophylactic after-treatment but als in other respects. From the very earliest days, FREUND (40), wh is generally recognised as the founder of the Roentgen therapy, ha kept on giving small daily doses; and he claims that he »in zah losen Fällen so gute Heilresultate erzielt, als mit Röntgenstrahle überhaupt erzielbar ist». In certain cases, however, he considers th intensive treatment to be the best. HOLZKNECHT (63), one of th most ardent advocates of the intensive method, is still recommendin »der einzeitigen Höchstdosis» in malignant tumors but regards it a unsuitable in other lesions. WERNER (178) has partly given up th massive doses and is now in favor of small daily doses for 12—1 days or more; particularly the palliative results are improved i this way. In 1926, HOLFELDER (58) writes of the intensive treat ment: »Dies chirurgische Behandlungsverfahren hat heute auc auf dem Gebiete der malignen Tumoren nur noch sehr wenig An hänger.» SCHINZ (155) states that it is only in the most sensitiv tumors that one may obtain complete destruction of tumor o single exposure to a dose that does not destroy the normal tissue In other cases one will have to give protracted continuous, or frac tional, radiation. WINTZ (180, 181) still recommends the intensiv method.

The method was never used as much anywhere as in Germany KNOX (72) recommends a cautious dosage with particular attentio to the general condition of the patient, which he is watching b frequent blood tests.

It is notably the French, though, who have been the pioneer with the less intensive treatments and have systematized thes methods of irradiation; the views of REGAUD have particularly bee widely recognised. REGAUD (142) emphasizes the importance o hitting all the cancer cells in their most sensitive state — the stag of cell division. For this purpose it is necessary to extend the radia tion time long enough for all the cells to pass this stage. There i no need, on the other hand, of any great intensity, as the cells ar comparatively easy to destroy while undergoing division. Th treatment of choice would therefore be a continuous weak radia tion, but at present such treatment can be given only with radium In the Roentgen therapy one is restricted to frequently repeate small doses.

As early as 1919, RUSS (149) has had some notions in this re pect. He was trying to estimate the time required for cell divisio

rom the time it takes tumor to double its size and from the pro-ortion between mitotic figures and other cells of tumor. From his estimate he would then decide on the method of radiation.

In 1923, NABIAS & FORESTIER (118) set up their karyokinetic ndex, whereby they mean the ratio between mitotic figures and ther cells. A low index would call for protracted irradiation; when ndex is high, the treatment should be shorter. To this HOLTHUSEN (61) says that the mitotic rate of the cells is at once disturbed on rradiation, the distribution of mitoses in the tissue being, besides, very irregular. LABORDE (84) also denies the practicability of a fairly dependable karyokinetic index.

In practice these principles are applied in various ways. REGAUD (142) gives radium rays for 6—15 days. In the Roentgen herapy he gives frequent small doses, whose total duration should not exceed 15—20 days, as otherwise the increased ray-resistance will assert itself. NABIAS & FORESTIER (118) recommend a distri-ution time of 6 days in index of 1 : 50 — 1 : 100, 15 days in index of 1 : 100 — 1 : 150, and still longer time when index is smaller. MALLET (98) gives 5—6 doses of X-rays a day for 6—30 days. MOR-AN (106) gives 2 doses a day for about 2 weeks. LACASSAGNE (86) gives 1 or 2 doses a day so as to give the calculated lethal dose within 4—10 days in superficial carcinomata, and within about 1 month in deeper tumors where several portals of entry are called for.

Every one of these authors thinks that he has seen better results rom his methods than obtained before, more marked effect on umor with less injury to the healthy tissues. I have not been able o find any comparative statistics of the brief and the protracted nethods of treatment. From the various statistics of radium-treated ancer of the uterus LABORDE (85) tries to find out whether the re-ults depend on the radiation time. She thinks, however, that de-inite conclusions are out of the question; there does not seem to e much difference. As to her own method, this author writes, in 927, that she had previously prolonged the radiation time, and that he now had returned to her old method of 3—6 days' radiation. his method, she claims, gives just as positive results as do the pro-racted radiations, and it does not give the very severe reactions net with in the very intensive treatments.

Other authors have attempted by other means to throw some ight upon this problem. ROUSSY, LABORDE & LEROUX (148) have reated two epitheliomata of scarce mitoses with the same dose; in ne case the dose was distributed over a brief period, in the other ver a long time. Both tumors disappeared very readily, showing o difference whatever — neither clinical nor histological. ROUSSY

(147) attaches greater importance to the condition of the strom than to the abundance of mitoses in the parenchyma of tumor.

Schwartz (159) treated one half of a large basal-cell epitheliom with 1 × 15 H and the other half with 3 × 5 H. He found no diffe rence in effect. In a case of cancer of the breast (160) he applie 1×18 H to one field, 3×6 H in 3 days to another field, and 2×10 H in 10 days to a third field. 2 months later there were recurrence in the first and the third field; while the second field was conspi cuous by the absence of recurrence.

At present it is hardly possibly from the clinical experiences t form any definite view as to which method gives the best results – the protracted radiation or the intensive treatment. Opinions diffe very much in this respect. Add to this, that the protracted radiatio unquestionably permits of a larger total dose without injuring the nor mal tissues, what makes it still more difficult to estimate the effect of equal doses given after the different methods.

All the previous methods are based on the view that the activ principle of radiation is acting only during the period of radiation However, one might also imagine that this agent would continu being active after the discontinuance of irradiation. Such actio may be taken for granted if some biochemical product of irradiatio is formed in the tissues. It is on this basis that Kingery (71) ha elaborated the saturation method.

If such an irradiation product is formed, its effect may be assume to be proportionate to its concentration. This effect will be greates right after the irradiation, whereupon it will decrease in proportio to the concentration, that is: most rapidly at first and more slow ly later on — in conformity with the law of mass reaction.

If one gives a single large dose, one will have a high concentra tion to begin with. But, it will soon fall off. And, with a certai large dose one will injure the normal tissues, as one will excee their tolerance limit, which may be assumed to lie at a definit concentration.

By giving a series of small doses it takes longer time to reac the high concentrations. The height of concentration will depen upon the size of the individual doses, their number, and the inte vals between their administration. By suitable adjustment of thes factors one may get to the very limit of tolerance without ove stepping it.

In order to obtain the greatest effect one must rapidly get u to the tolerance limit and then keep the concentration near th point: the tissues must be kept saturated for some time.

Kingery (71) was using a rather soft and unfiltered ray quality

and he has constructed some curves wherefrom he could read the residual effect at any time after the irradiation and, consequently, also the dose required for regaining the saturation point. 2 weeks after he had given an erythema dose he could repeat it without any injury to the skin. The curve must therefore have dropped to zero in the 2 weeks. He then tried how soon after the first dose he could give 75 % HED, and he found that this could be done on the 7th day. In the same way he found that he could give 50 % HED after $3^1/_2$ days. From these points he constructed his curve which gives the residual effect at any time. Later experiments have proved the accuracy of this curve.

KINGERY has used this method only in the surface therapy. In practice he gives 50 % doses as a rule. He does not like to use shorter intervals, as the individual differences in sensitivity always imply some risk of cumulative effect. I have not been able to find any report by KINGERY on clinical results. HAZEN & WHITMORE (51) have treated two cases of epithelioma after Kingery's method; and they got a very marked skin reaction.

PFAHLER (136) has introduced this method in the deep therapy, and he said in his paper before the First International Congress on Radiology, London, 1925, that he has not obtained such good results with any other method, in spite of his wide experience with all the other methods of Roentgen therapy.

PFAHLER has worked out 3 different curves of the residual effect of different ray qualities. With the hard ray qualities the effect is decreasing more slowly, of course, than with the softer ones. He further emphasizes that it must not be assumed that small doses can be kept up indefinitely, even though the tolerance limit is not reached. Such doses may also be followed by atrophy, endarteritis, fate degenerative processes, and even necrosis. But, if the saturation is kept up long enough for all the cancer cells to pass through the most sensitive stage, mitosis, there is a better chance of a favorable result. The method has the advantages of the single massive dose treatment as well as of the protracted radiation after Regaud's method. The author has given up the single massive dose methods except in lesions that can be safely overdosed. He says in conclusion: »It is our duty in the treatment of deep seated malignant disease to administer to the tumor area the largest dose (an erythema dose, saturation), that is practical within a short time, and without damage to surrounding essential normal tissue or organs. This dose, when divided, should be given on successive days, so as to avoid the development of a decreased sensibility of the malignant cells. We should then continue this saturation over a period long

enough to destroy all carcinoma cells during their process of div
sion, and yet not continue it long enough to obtain a cumulative
destructive effect upon normal tissues, nor a late degenerativ
effect. We, therefore, approximate »saturation» as quickly as po
sible and continue it over a period of ten to fourteen days.»

In another paper, PFAHLER (137) reports some cases of metast
tic tumors of the bones that have been treated after this metho
The results are particularly good.

*Still, it is all too early yet to form a judgment of the clinical valu
of the saturation method.*

III. ON THE EFFECT OF LOCAL IRRADIATION UPON TRANSPLANTED TUMORS IN ANIMALS ON SINGLE EXPOSURE TO DIFFERENT DOSES.

A. PREVIOUS INVESTIGATIONS

The fundamental investigations, at the beginning of this century, into the transplantability of malignant tumors opened wide vistas of the practicability of studying the influence of various factors on the tumors. For now it was possible to obtain a large and homogeneous material for comparative examination.

Among the first to carry out such studies was C. O. Jensen (65) whose pioneering work in this field is known all over the world. Later investigators in every country have continued the work along these lines.

About a dozen years later another Danish investigator, Johannes Fibiger (38) accomplished the experimental production of malignant tumors in animals. His epoch-making work on the spiroptera cancer has made his name world-famous. Subsequent experimental studies however, have chiefly been carried out on tumors produced through application of tar. These experimentally produced tumors resemble the spontaneous tumors more closely than do the transplants. But, it is yet very difficult to produce a large and homogeneous material in this way; and it is also hard to obtain a sufficiently large number of spontaneous malignant tumors in animals.

Most experimental work on the influence of radium and Roentgen rays upon malignant tumors is therefore done with transplanted tumors. Many of these studies are based on radiation treatment of tumor tissue *in vitro* or *in situ*, with following transplantation for indication of the effect.

Regarding these investigations I may refer to the works by C. Krebs (78, 79, 80) and merely state that the great majority of authors have found that it was feasible indeed to destroy the tumor cells, but that the dose required for that purpose was far

in excess of any therapeutic dose (CONTAMIN, 28, CLUNET, 27 MEISEN WESTERGAARD, 102, WEDD. & RUSS, 177, v. WASSERMANN 176, PENTIMALLI, 129, 130, HILL, MORTON & WITHERBEE, 52, WOOI & PRIME, 183, RUSS, CHAMBERS & SCOTT, 150, KEYSSER, 70 MURPHY, MAISIN & STURM, 111, KOK, 74, KREBS, 80).

A few of these authors (MEISEN, WASSERMANN) have found th lethal dose to come close to the therapeutic dose, but subsequen extensive investigations seem conclusively to have demonstrate that the lethal dose for most tumors, at any rate, is considerably higher — and not a great deal less than the dose which destroys th normal tissues. (PRIME, 138, MURPHY, MAISIN & STURM, 111) Several of these authors have further shown that the percentage o takes and rate of growth decrease on increase of dose.

As to a number of investigations on preceding radiation treat ment of the animal and following transplantation into the treate field I may likewise refer to KREBS. From the findings of CLUNET (27), FRANKL & KIMBALL (39), MURPHY and co-workers (110 96, 111, 119), KOK (74), BECK & ENGEL (13), RUSS & SCOTT (153 and KREBS (80) it must be taken as established that preceding irra diation is capable of lowering the percentage of takes and of retardin the rate of growth in transplanted tumors. NAKAHARA (119) an VORLAENDER (173) have further shown that the histological changes which occur in tumor under these circumstances, are identical wit the changes which follow direct irradiation of tumor *in situ*.

These various experiments indicate very strongly that, in radia tion treatment of malignant tumors, the direct effect upon th tumor cells is at any rate only one aspect of the process, and tha the reaction of the surrounding tissues is of great — if not decisiv — importance.

Other investigators have experimented with direct irradiatio of transplanted animal tumors *in situ* some shorter or longer tim after the transplantation, and they have studied the effect on th growth of the tumors.

As early as 1901, LOEB (97) treated a transplanted sarcoma in ra with repeated X-ray doses. He found no definite effect, however but he thinks that the doses were too small.

APOLANT (8) has treated various mouse carcinomata, which showe several instances of spontaneous recovery, with 20 mgm. of radiu in mica capsule, and he made most of the tumors disappear. H gave several applications of different duration and at differen intervals. The doses appear to have been rather large, as epilatio occurred early in the treatment and, in most instances, was fol lowed by erosion and scab formation, in a few by ulceration.

Bashford, Murray & Cramer (12) treated Jensen's mouse carcinoma with radium and obtained a diminution of the tumors. They could not demonstrate any relation between the size of dose and the degree of effect.

Meisen Westergaard (102) likewise worked with Jensen's mouse carcinoma. On local treatment of the developed tumors with various X-ray doses, distributed over various lengths of time, he obtained a transitory inhibition of growth and increased consistency in some of the cases and disappearance of tumor in one case, which, however — as he remarks himself — may occur spontaneously. No prolongation of life could be demonstrated.

Contamin (28) treated an adenocarcinoma of mice with soft X-rays; the dosage is not given. In animals with smaller tumors, transplanted 15—20 days before, the tumors disappeared, and the animals recovered. In animals with larger tumors, the tumors diminished, but the animals died; the author thinks that death here is due to intoxication from absorbed autolytic products of tumor tissue. In animals where tumor had been transplanted 2—3 months before but had stopped growing when as large as a nut, the tumor did not disappear. From this he draws the conclusion that tumor is affected the more readily, the younger and the stronger proliferating it is. As to the cause of death in the middle group, it seems more reasonable to assume that the animals died from the radiation, as the size of tumor made it necessary to expose a larger area. The absorption in this group is uncertain, too, as the size of tumor is estimated from changes in the weight of the animal. The animal itself has very likely lost considerable weight as a systemic effect of the radiation.

Clunet (27) has treated some carcinomata of mice with 30 H and observed a retardation of growth in some of them. In this report, however, as in most other works from that time, the author is particularly interested in the histological changes.

Menten (103) found noticeable effect from radium treatment only in Jensen's mouse carcinoma; two other tumor types were not affected.

Levy-Dorn (94) found that mouse carcinomata and mouse sarcomata were likely to subside on treatment with X-ray doses not less than 4 HED. Some of these tumors, however, were very refractory to treatment.

Caan (21) made several mouse carcinomata and rat sarcomata disappear by intratumoral injection of thorium chloride solution.

Hirschfeld & Meidner (53) treated sarcomata of mice and rats with injection of thorium X-aluminum hydroxide and obtained

a marked necrotic effect, in some instances with shrinkage and decay.

Wassermann (176) could produce only superficial necrosis of tumor by treating it several days with 50 mgm. of mesothorium, while it continued to grow.

Pentimalli (130) found no effect on Ehrlich's mouse carcinoma from radiation treatment with mesothorium, not even with doses larger than the therapeutic. Nor did he find any effect on radiation shortly after transplantation. Many of his animals died from the treatment, evidently from a severe general effect of the irradiation.

Paul Lazarus (91) treated 2 mice with 100 mgm. of strongly filtered mesothorium at a distance of 1—2 cm. and 2 other mice with the same dose at a distance of 20—25 cm. In the first two mice the tumors decreased much in size, in the last two the tumors grew excessively.

Keysser (70) has treated intramuscular carcino-sarcomata of mice. The tumors decreased in size, some even disappeared altogether, on treatment with 2—4 HED of X-rays of varying hardness. He further showed that tumors, which had diminished on treatment with such doses of Roentgen rays, were capable of growing anew — although somewhat slowly — when transplanted into other animals several days after the radiation treatment.

Murphy, Maisin & Sturm (111) treated autografts from cancer of mice with an erythema dose of soft X-rays on the 10th day after transplantation. The tumors disappeared in 76 % of the mice, while non-treated autografts grew in 96 % of the mice.

The purpose of all the above-mentioned experiments has principally been merely to find out if it is possible to affect these animal tumors by means of radiation, or to investigate the histological changes after radiation. Most of the experiments were made with only few animals.

Russ, Chambers & Scott (150) were the first to make a more thorough study on the effect of different doses. They exposed sarcoma of rats to local treatment with β- and γ-rays from 20 mgm. of radium bromide distributed over an area of 4 cm . With very small doses the growth was rather more rapid than in the control animals; with larger doses there was an inhibition proportionate to the dose. Repetition of the treatment once a week gave similar results. The stimulating dose was below 1/50 of the lethal dose for the tumor tissue. The highest dose employed was 10 min. once a week. The lethal dose for tumor tissue *in vitro* was 40 min.

Nather & Schinz (123) have treated carcinoma of mice with

single doses ranging from 1 % HED to 375 % unfiltered and 100 % filtered with 3 mm. Al. None of the tumors disappeared, but growth was inhibited by the large doses. No stimulating, growth-promoting effect could be demonstrated. The experiments loose something in significance by the facts that each group comprises only 3 animals, and that many of the animals died from the radiation treatment.

Caspari (26) found that in local treatment of mouse carcinoma the effective doses are much higher than the doses for man. He found it necessary to give at least 1½ HED, and he has never seen recovery after doses less than 3½ HED. He has never seen any increase in rate of growth after doses up to 1 HED. Improvements was some times seen after doses of 2½—7 HED, whilst other tumors appeared to grow particularly fast after these doses. The latter effect is due, the author thinks, to necrohormones produced through irradiation.

A larger series of experiments concerning the effect of local X-ray treatment with different doses has been reported from Freiburg by Kok & Vorlaender (73, 74, 76). These authors worked with Ehrlich's mouse carcinoma, used hard rays filtered through ½ mm. Cu and determined the size of dose with the ionto-quantimeter. They applied the radiation partly to a small area corresponding to the size of tumor, partly to a slightly larger area so that the surroundings of tumor were also treated. Their largest dose was 490 e — 1 HED for man is 170 e, the epilation dose for mouse is 250 e. It was found that in local treatment the best retrogressive changes were obtained with 200—225 e, that is a dose which is a little less than the epilation dose for mouse. Larger doses gave poorer results. The effect was better on irradiation of a large area than of a small area. — The experiments were repeated with irradiation shortly after transplantation, and similar results were obtained. In this series, however, doses as low as 100 e were also found to be active, although to a less extent. — Local irradiation of the animal prior to transplantation showed the same relation between size of dose and effect. — The authors interpret their findings in this manner, that the large doses injure the surroundings of tumor so much that the therapeutic result is less favorable, in spite of a possible stronger direct effect upon the cancer cells.

On summing up all these radiation experiments it is found that most transplanted mouse and rat tumors have proved rather hard to affect with radiation, and that the requisite dose is much higher than the dose for human beings. Not many authors have given the ratio

of effects obtained with different doses, but most authors appear to hav got the best effect with the largest doses. Exceptions hereto are Ko & Vorlaender, who found a decrease of effect in the very larg doses, and, to some extent, Caspari, who has seen both good an bad results from such doses. Kok & Vorlaender's experiments on the other hand, are considerably more extensive than most o the others, and their maximal dose appears also to be higher. A re petition of their experiments might therefore prove to be of signi ficance.

B. OWN INVESTIGATIONS

1. *Material, Technique and General Arrangement of Experiments*

Animals:

The experiments are carried out on white mice with transplan ted tumors. As a rule the animals were pretty young, 3—4 months all were purchased from the same breeder. The weight of the ani mals has varied somewhat; most of them weighed 15—22 grams During the experimental period the animals were kept in smal cages, each containing up to 6 mice. Their food has been whea bread and rye bread, soaked in water and squeezed.

The outer conditions and the food were the same for all th animals. Within the different groups of each experiment the ani mals have all been of the same sex and — as far as possible — o the same weight. None of the animals had previously been use in transplantation or radiation experiments. Pregnant animals wer used in but very few instances.

The precautions, I think, furnish the best possible proof agains differences in the resistance of the animals that arise from race age, sex (pregnancy, lactation), food and other outer conditions (Apolant, 9). By never using any animal which had been use in any experiment before I have further avoided all changes i resistance from previous transplantation (Jensen, 65, Ehrlich 34).

There has been no epidemic of any kind among the experimenta animals; and very few of the nearly 1,600 mice employed have die from causes other than what were naturally attributable to th size of tumor or to the treatment.

Tumors:

The experiments are made with two different transplantabl tumors, an adenocarcinoma and a sarcoma. They were placed a

my disposal by C. KREBS, and they are the same tumors as he has used himself in his studies on the effect of radiation on tumor tissue and on the organism (79, 80). As to the details about these tumors I may refer to his works and merely mention that the carcinoma comes from a spontaneous tumor in a mouse, probably a cancer of the breast, while the sarcoma is produced by tar application.

Both tumors show their malignant character by their

1) invasive growth,
2) production of metastases, and
3) transplantability.

Both tumors have been transplanted in several generations before the experiments were started, and they have not changed in character in the two years the experiments have been going on. Both tumors have been kept growing in a pure side-line, so that further transplantation in no instance is made from an animal in which the tumor had been treated in any way at all.

The microscopic findings in both tumors are described in detail and discussed in the works by KREBS, wherefrom the following description is taken:

Carcinoma (Tabula I):

On microscopic examination the tumor is found to have an alveolar structure, rich in epithelial cells, with a rather sparse connective tissue stroma. The tumor cells form larger and smaller islands and anastomosing strands. Often the cell groups are solid, but many areas show definite glandular formations of round or irregular shape and with the epithelial cells arranged in one or several layers. The tumor cells are small, cuboidal or round, with oval or round nuclei, whose chromatin has a peripheral position so the nuclei appear vesicular. The cytoplasm is scant. The cells are of epithelial character, and they show no sign of keratinization; but few mitotic figures are seen. No basal membrane can be made out. In many places the stroma is so edematous that the structures can hardly be seen. Nevertheless, the alveolar arrangement of the epithelial cells is quite distinct. There are smaller and larger areas of necrosis and more or less dense infiltration with leucocytes. The tumor tissue shows definite invasive growth.

Metastases in the lungs show the same microscopic features as the primary tumor. Many of the blood vessels are filled with tumor masses, which sometimes form emboli.

Sarcoma (Tabula II):

Microscopic examination shows a remarkably cellular tumor, consisting of polymorphous cells and an exceedingly sparse stroma.

There is only a very slight indication of the cells being arranged in strands. In a few areas the spindle-shaped cells are predominating. Most of the cells are polyhedral or round, generally large, though varying in size. The cytoplasm is scanty. The nuclei are large, oval, round, or irregular, with a deeply staining chromatin network of varying pattern. Scattered throughout tumor — now and then in great numbers — are very large cells with very large nuclei, or with several nuclei like small and large giant-cells. The shape of the cells as well as the nuclei are irregular. Many mitotic figures are seen. The inconspicuous stroma is composed of small blood vessels and fine collagen fibrils, which are diffusely distributed in direct contact whith the tumor cells. There is not even a suggestion of any alveolar arrangement of the stroma. Here and there in the tumor tissue are striated muscle cells, which indicate the marked capacity of tumor for invasive growth; many of these muscle cells show definite degeneration. Finally, there are more or less extensive areas of necrosis with kariolysis and fading of structures.

Metastases in the lungs show the same features as the primary tumor. Neither the metastases nor the primary tumors show any sign whatever of keratinization.

Transplantation technique:

In the preliminary experiments the same transplantation technique was employed as KREBS has used and described in his reports (78, 80). For this reason I shall outline it but briefly. The mother animal is killed with ether; tumor is removed sterilly, cut in pin-head-sized pieces and kept in a sterile Petri dish with a few drops of sterile physiologic salt solution. The experimental animals, which have already been epilated over a small area on the lower part of abdomen, are anesthetized with ether and tied to a board; the epilated area is painted with tincture of iodine, and an incision — about 1 cm. in length — is made through the skin. From this incision a blunt instrument is pushed up into each axilla, wherein a small piece of tumor tissue now is introduced by means of a pair of fine pincers. The wound is then closed with a catgut suture and collodion. The whole operation is performed with sterile instruments.

With this method transplants of the sarcoma take in ca. 100 %, of the carcinoma in 40—60 %. The method has been used in the experiments with sarcoma and radiation on 2'—3' day after transplantation, partly universal irradiation, partly local treatment with doses up to 6 S.—N.

There were difficulties, however, which necessitated a change

in the technique. It was found that the animals could hardly stand a dose of more than 6 S.—N. on that region of thorax and upper abdomen which had to be treated. The great majority of the animals that had got a larger dose died in about 2 weeks. And it was almost impossible to produce any radiation effect upon the sarcomata that were transplanted in this way, probably on account of their extraordinarily rapid and invasive growth.

To get over the first difficulty I went on to implant the transplants in the thighs, in the skin fold extending downwards from the base of the tail. An advantage of this procedure was that the animals could stand a considerably larger local dose, probably because it leaves out the irradiation of large parts of the abdomen without implying the risk of failing to treat a part of the tumor or the transplantation site. Experiments by WARREN & WHIPPLE (175) and MARTIN & ROGERS (100) suggest very strongly that changes in the small intestines are the cause of death after radiation. FABRICIUS-MØLLER (37), too, has found some changes in the intestines, although he does not take them to be the cause of death. At the same time I proceeded, for practical reasons, to transplantation with cannula instead of through incision, and I have used this method in the majority of my experiments, that is: in all the experiments with the carcinoma and in the experiments on local treatment of the sarcoma with doses above 6 S.—N. on the 2'—3' day after transplantation.

The method then has been as follows: Tumor is removed from the mother animal, cut in pieces and kept in a sterile Petri dish, as outlined before. The experimental animals, which, beforehand, are epilated on a small area at the base of the tail, are held at the neck and the tail by an assistant. The epilated area is painted with tincture of iodine. A small piece of tumor is placed at the tip of a fairly wide cannula (lumbar cannula), and the cannula is introduced through the iodine-treated skin down into the subcutaneous tissue of the mentioned skin fold, where the tumor tissue is pushed out with the stylet. The process is repeated on the other side.

In most instances the tumor has developed at the intended site and subcutaneously; a few times it has appeared on the back near the skin puncture; and in a very few instances it developed underneath the fascia, within the muscle.

With this method growth took place on an average in about 70 % of the carcinoma transplants and in 90—100 % of the sarcoma transplants. In the carcinoma transplants growth usually results within 3—4 weeks in a round or oval tumor, measuring 5—8 mm. in diameter, which keeps on growing steadily till the animal dies,

on an average 10 weeks after transplantation. Metastases in the lungs are met with, but they are rare in the control animals and somewhat more frequent in the treated animals, possibly because the life-time of the latter is prolonged. — The sarcoma transplant shows a tumor of similar size at the end of 1 week, and the animals die about 4 weeks after transplantation. Here metastases in the lungs are frequent.

As the sarcoma grew so very rapidly, I have tried to make it grow more slowly by another method. C. O. JENSEN (65) has shown that the transplantability of a tumor can be destroyed on grinding with kieselguhr; and perhaps, I thought, one might succeed in lowering the rate of growth too by this means. My method was as follows. The tumor is ground in a sterile mortar with sterile sand or kieselguhr and sterile sodium chloride solution. On standing, most of the sand or kieselguhr will settle to the bottom, and the supernatant fluid, which on microscopic examination is found to contain tumor cells, is injected subcutaneously in the thighs by means of a Record syringe. In some instances this method really resulted in tumors that developed and grew more slowly, but it was connected with so many difficulties that I gave it up. For one thing, it took so many animals, as the percentage of growth was lowered very considerably and, then, several series of animals showed abscess formation so that all the animals had to be discarded; besides, when too much sand or kieselguhr was injected together with the suspension of tumor cells, it set up a tissue reaction with infiltration at the site of injection that was very hard to distinguish from a beginning tumor. The only series of experiments that is carried out after this method is the experiments with irradiation of the sarcoma after the first appearance of tumor.

It is a well-known fact that transplanted tumors may disappear spontaneously. The frequency of this retrogressive process varies in the different types of tumor; in some forms it is a very common occurrence (APOLANT, 9). In my experiments I have seen 2 out of 68 carcinomata disappear spontaneously. I have seen only one sarcoma disappear altogether; another sarcoma, in a pregnant mouse, grew at first and then subsided very markedly after delivery (during the lactation), only to grow again a couple of months later.

Relative to both tumors the various animals have shown some individual difference in the rate of development and growth of tumor. Thus it was evident that the tumor made its first appearance sooner and grew more rapidly in young animals than in older ones, which has also been observed by many cancer-investigators (APOLANT, 9).

Radiation technique:

The radiation treatment has been given with a twin-coil apparatus (Neo-symmetric) from Reiniger, Gebbert & Schall and a Coolidge tube from Müller (Hamburg). The strength of the current used was 2.8 MA, the tension at the tube was 220 KV max. measured by the spark gap between spheres of 12.5 cm. in diameter, as the spark-length was 10.7 cm. The Voltmeter showed 142 KV eff., measured on the primary current. Considering the type of apparatus, these technical figures should be judged with some reservation, but they show at any rate that the ray quality is rather hard. For filter was used 5 mm. Al in all the experiments. Kok & Vorlaender used ½ mm. Cu. It is for practical reasons that I did not use the same filter, as the radiation time with copper filter would amount to several hours in the most protracted applications. I therefore decided on 5 mm. Al so as yet to get a fairly hard ray quality together with a practicable radiation time. In every experiment the distance from focus to animal was 23 cm.

Under these circumstances 1 HED for man takes 21 minutes, when the field is 10 × 10 cm. In daily practice in the clinic the dosage is done by time, but the doses are calculated in Sabouraud-Noiré units. It is found that with the factors mentioned and a focus distance of 23 cm. 1 S.—N. takes 6 min. 1 HED of the mentioned ray quality corresponds to 3½ S.—N.

For local treatment the animals were tied down, under anesthesia, in ventral position on a board, 2 at a time, with loops of coton yarn around the extremities and the tail fixed with adhesive tape. In this way the animals were unable to move. For protecting-screen I have used 1 mm. lead plates, bent so as to fit the bodies, 2 plates to each animal, one covering the forepart of the body, the other plate covering the tail and distal part of lower extremities, both plates fixed with adhesive tape. Between the two plates was a gap of 2 cm. so the animals were treated on a 2 cm. wide zone across the lower part of the back, the base of the tail and the upper part of the lower extremities.

I have tested the lead plates I used for protecting-screen to see if any rays passed through, and with a Siemens dosimeter I found that the rays passing through amount to 4 % at the most; this means that with the largest single doses, where 10 S.—N. were given locally, the rest of the animal got 0.4 S.—N. at the most. As doses up to this size have shown no effect on universal irradition, I take it for granted that this factor has not had any particular influence on the result.

As well known, the total radiation effect — the effect of bot primary and secondary radiation — is less in a small field than i a large field. By dosimetry on paraffin bloc I naturally found somewhat smaller value in the field for mouse-irradiation than in field measuring 10 × 10 cm. No difference, on the other hand, wa found on direct dosimetry on the operating table (the board).

In animal irradiation, therefore, the effectivity of a given dos is undoubtedly smaller than that of the same dose applied to larger field in a human being. Considering the small size of the an mal, ca. 1 cm. in ant.-post. diameter, and the small field, the secon dary radiation does not amount to much (WINTZ, 181). In th present investigation, however, the absolute dose is of minor impor tance, as I have aimed merely to throw some light on the relativ conditions. I do therefore not attach any great importance to th dosage in S.—N. and HED, but I would like to emphasize the stan dards one has in the effects on the mouse, the epilation dose in loca treatment and the lethal dose in universal treatment. Here, too we meet with some individual variation, as young mice in particula are more readily affected than older mice. With the technique em ployed epilation is often produced by 5 S.—N., usually by 6 S.—N and never by less than 5 S.—N. In universal irradiation with th above-mentioned factors the lethal dose is around 3½ S.—N., tha is about 1 HED for man.

These standards, too, form the best base for comparison wit doses in earlier investigations of similar nature. They indicat fairly exactly the amount of rays that has taken effect in the ani mal, and in this way they obviate the difficulties arising from th size of field and the more or less dependable physical standards used

General arrangement of experiments:

Some of the animals are treated after the development of tumor others are treated 2—3 days after the transplantation — befor any tumor could be palpated.

The first group has been handled as follows: Transplantatio is performed on a certain number of animals of same sex that sta in a large cage, until there is a sufficient number of animals wit tumors large enough for the planned experiment. I have preferre tumors that were 5—7 mm. in diameter; only few have been smalle very few have been larger. I did not mind, though, that tumor o one side was undersized if only the other came up to the standard Individual differences have made it impossible to get tumors tha were of exactly same size. When the time came, I have picked ou the animals that were suitable for treatment and divided them in a

many groups as the experiment would comprise, in such a way that the animals in the different groups as far as possible were of the same size and had tumors of equal size. The weight of the animals was put down, the size of their tumors was drawn in the protocol, and the animals were marked by cuts in the ears, whereafter the treatment was instituted. The first 4 weeks the animals were examined every week, thereafter every other week, and the size of tumor was drawn in the protocol as on the first examination.

Certain difficulties were encountered in estimating the size of the tumors. The tumors, however, that were located immediately under the skin are comparatively easy to palpate and measure. I have measured them with a caliper-square, seizing the tumor between its cross-heads. I have carefully palpated the tumors at the same time, and sometimes, particularly in judging the very smallest tumors, I think, one may get the most accurate estimate by palpation alone. After some practice I feel that I have attained a fair degree of accuracy; and it is my opinion, that this combination of judging and measuring gives a more accurate estimate than does judgment alone. Though there be some error in the absolute values, the errors in the relative measures are undoubtedly smaller. The third dimension of the tumors cannot be ascertained in this manner; and I can see no way of doing it when one does not want to kill the animals. But, such a delineation of tumor as a silhouette has also been used in most works of similar nature all over the world.

All the animals, whose tumors did not disappear, were allowed to die a natural death. Autopsy has been performed on all of them, with examination for metastases; pieces of tumor and metastases were removed and preserved for eventual microscopic examination. The animals, whose tumors did disappear, were allowed to live 6 months longer, except in particular instances that are mentioned especially. Then they were killed, and autopsy was performed with careful examination of the implantation site and the lungs in particular. Microscopic examination was not made where no macroscopic abnormality was found. If, however, there was anything suggestive of tumor tissue, it was examined microscopically — in some instances in serial sections. At the time when these animals were killed, most of the mice with persisting tumors had already been dead a good while.

I have seen recurrency only in 3 cases without any palpable tumor for more than 2 weeks. All 3 were carcinomata; there had been no palpable tumor for 6 weeks in one of these cases and not for 2 months in the two others. There has been no recurrency after a longer period.

The great majority of the animals whose tumors disappeared, have shown no macroscopic abnormality. A few of them have shown a slight whitish or brownish thickening at the site of tumor, but no tumor tissue could be demonstrated on microscopic examination. It was granulation tissue or localized hyperplasia of the connective tissue; foreign-body »tubercles» were demonstrated in one single animal that was inoculated with a tumor that had been ground with sand.

Nor has there been any sign of metastasis in these animals. In 2 animals the lungs presented some metastasis-resembling nodules. In one of the cases microscopic examination proved the nodules to be pneumonic processes; in the other case it was a tumor of the very same structure as the pulmonary adenoma, described by several authors, which is a non-infrequent spontaneous tumor in mouse (Apolant, 10, Murray, 117, Murphy & Sturm, 115, Fibiger & Møller, 38).

Whether the animals in which the tumors have disappeared are really cured, that is a question I cannot answer conclusively. The question is not of decisive importance to this study, whose purpose is to elucidate the relative conditions in the different radiation methods. Still, the clinical course of the lesion as well as the autopsy findings indicate that the animals are cured.

In the other experimental series, where the radiation treatment was given 2—3 days after the transplantation, the animals were segregated in the small cages on the day of transplantation so that the members of the different groups were of approximately same weight. They were examined every week as long as tumor might be expected to appear, and less frequently later on.

The animals in which the tumor developed were allowed to live, until they died a natural death. The animals without any tumor were killed after different lengths of time that will be mentioned in connection with the different experiments; but, none of these animals was killed before all the others in the same group had died from their tumors. When an animal died from other causes so early that the tumors were very small or questionable, they were always examined microscopically before they were recorded as positive growth. Animals that died before tumor could have developed are not included in the result.

Beside the mentioned examinations, microscopic examination was made of all the tumors used for transplantation. I have made additional microscopic examination of numerous tumors and metastases, chosen by chance, and I have always found the features one

would expect. Most of the sections were stained with glychæmalin-eosin.

In contrast with KOK & VORLAENDER, who have laid much stress upon the microscopic appearance of tumor and its surroundings in their estimate of the results, I have gone after the macroscopic findings and conspicuous conditions. The three criteria I have been relying on for indicators of any favorable effect from the radiation treatment are:

1. Tumor's rate of growth.
2. Number of tumors that have disappeared, or percentage of takes.
3. Average of life-time in animals with persistent tumors.

In experiments where the tumor had not appeared at the beginning of treatment, the incubation time — that is, the time between transplantation and manifestation of tumor — is also taken into account.

2. *Experiments with the Carcinoma. Irradiation after Manifestation of Tumor.*

This series comprises 5 groups of animals: 1 group of non-treated control animals and 4 groups that were treated, respectively, with doses of 2, 5, 7, and 10 S.—N. The collective series is composed of several smaller series. In each of the smaller series care was taken as far as possible to have each group comprise the same number of animals of the same weight and with tumors of equal size. A few of the animals — particularly in the groups that were treated with the large doses — died before their time, probably mainly from the treatment, as the field has extended too far up on the abdomen; and this has brought about that the various groups do not comprise quite the same number of animals that could be included in the total accounts. To counterbalance the relatively heavy loss of animals in the groups treated with the large doses, I have a few times included a few additional animals in these groups at the beginning of the experiment. This step can hardly have any influence on the total outcome, as the various sub-groups differ very little from each other.

Schema I gives the outline of the tumors, reduced to $^1/_4$ natural size (linear reduction), at the beginning of the experiment and after 4, 8, and 12 weeks. There is a definite gradual decrease in the rate of growth as the dose is increased up to 10 S.—N. Even the smallest dose employed, 2 S.—N., gives a considerable inhibition with no indication whatever of a more rapid growth.

Table I gives the number of animals in which tumor disappeared, and the average life-time of the animals that died with tumors.

Table I.

	Tumor disappeared in	Average life-time (with persisting tumor.)
Controls	1 out of 25 — 4 %	46.3 days
2 S.-N.	1 » » 21 — 4.8 »	63.5 »
5 S.-N.	2 » » 22 — 9.1 »	76.8 »
7 S.-N.	5 » » 23 — 21.7 »	93.2 »
10 S.-N.	8 » » 23 — 34.8 »	115.7 »

As to the disappearance of tumor, definite effect is first obtained with 7 S.—N.; and this effect is still more pronounced on treatment with a dose of 10 S.—N. The tumor disappeared spontaneously in one of the control animals. This has happened in only 2 out of 68 control animals with carcinoma.

The average life-time is seen to increase quite gradually with the increase in dose.

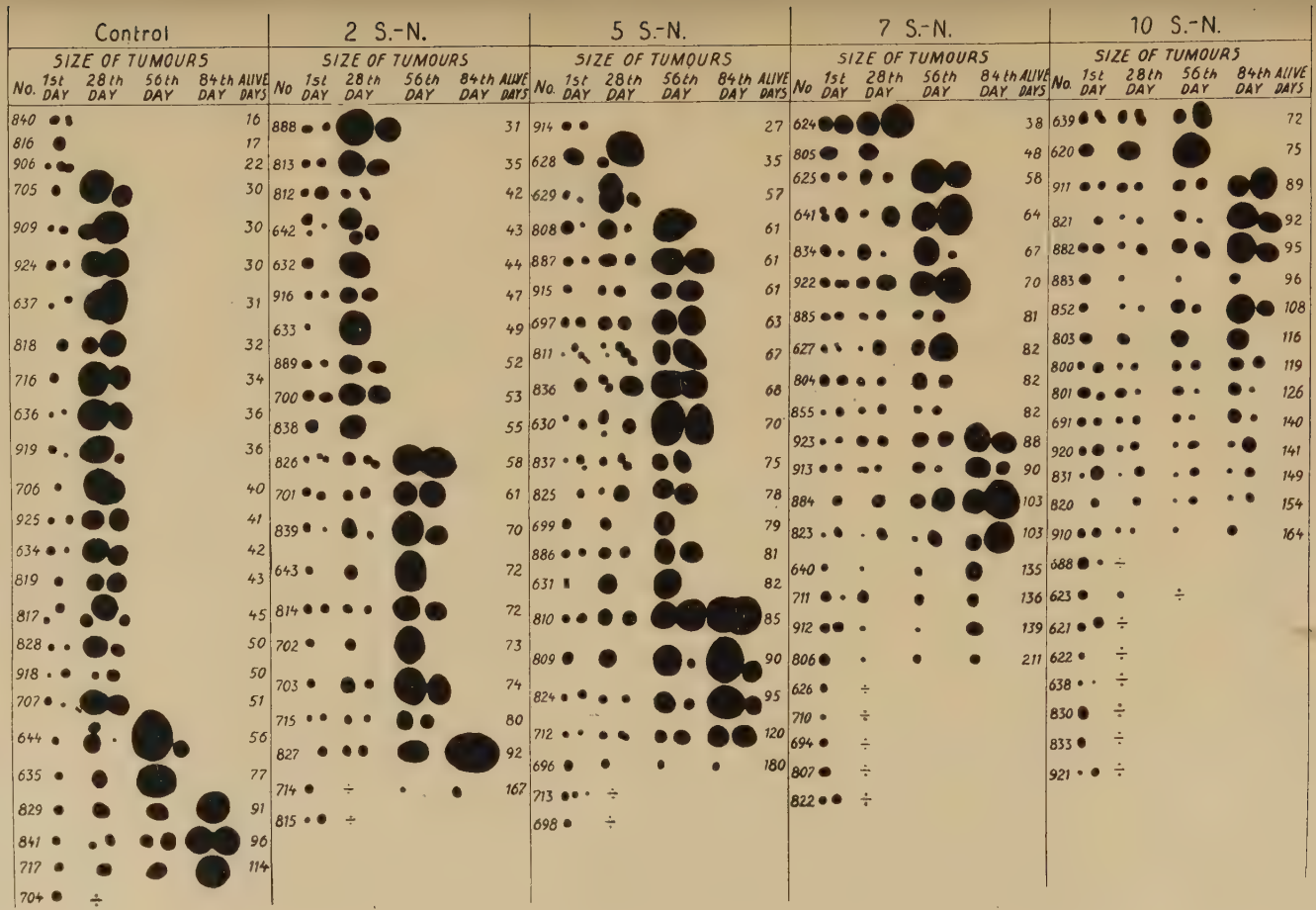
All the three criteria show then: an increase in effect when the dose is increased — all the way up to 10 S.—N.

5 S.—N. gave often epilation, 7 always. 10 S.—N. gave epilation, often excoriation and crust formation in the early stages and later, after some months, contraction that caused the hind-legs to be drawn in toward the base of the tail, so the animal had difficulty in moving about and walked with a marked adduction of the lower extremities. The otherwise thin and soft skin fold that runs from the base of the tail to both sides became thick and firm; this fold as well as the base of the tail were often the site of ulcerations. I am mentioning these findings because they show that there was really given doses which cause rather marked injury to the normal tissues. Such changes were also observed after treatment with 7 S.—N., but not so often and not so pronounced.

The results cover 114 animals altogether. I shall briefly mention the animals that are left out from the schema and the calculations.

No. 689 and 802 died 6 and 8 days, respectively, after exposure to 10 S.—N.; the latter had intestinal hemorrhages. No. 708, 690, and 709 died 18, 19, and 20 days after treatment with 10 S.—N.; the tumor was steadily diminishing in these 3 cases. No. 695, 693 and 692 died 4, 4, and 9 days after treatment with 7 S.—N., the last one with diminishing tumor. No. 917 died 37 days after treatment with 2 S.—N.; it had a very small tumor that kept on diminishing until death.

14 out of 17 animals, whose tumors disappeared, lived 6 months or longer after the tumors were impalpable. 1 (treated with 10 S.—N.



Schema I. Carcinoma. Local irradiation after manifestation of tumor. Tumors in $^1/_4$ natural size.

died 3 months after the disappearance of tumor; 1 (treated with 2 S.—N.) died 1 month after the disappearance of tumor; and 1 — control animal —died 5½ months after the spontaneous disappearance of tumor. Autopsy revealed no macroscopic evidence of tumor or metastases.

3. *Experiments with the Carcinoma. Irradiation Shortly after Transplantation.*

Doses of 2, 6, and 10 S.—N. were used in these experiments. The total outcome of the experiments is the addition of the results from 3 smaller series, each of which gave the same result as the total. Each of the smaller series was made up by a group of 12 control animals and 3 groups of treated animals — 6 in each group. As far as possible the animals in the various groups were of the same size. The treatment was given on the 2′ or 3′ day after transplantation.

Schema II gives the outline of the tumors in ¼ natural size 5 weeks after transplantation. The sign : designates the absence of a palpable tumor; : (+) means a later appearance of tumor; (+:-) after tumor signifies that tumor later disappeared; (died) means that the animal died before the expiration of 5 weeks without any tumor.

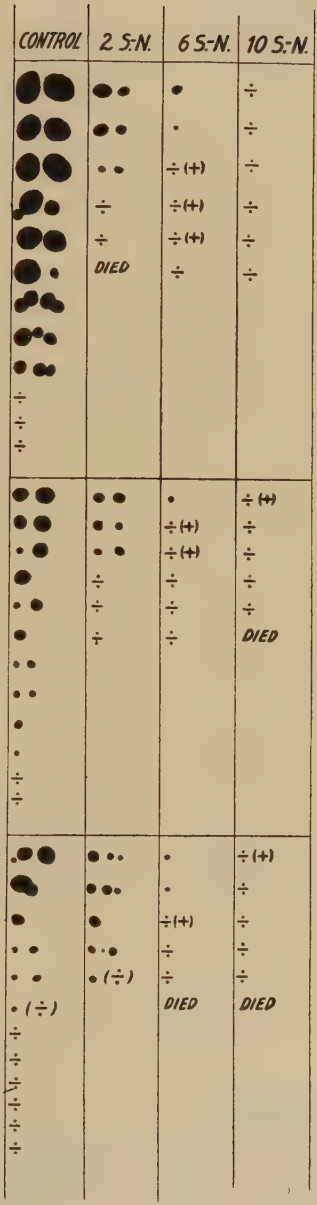
The various groups differ conspicuously in the size of tumors, and the greatest effect is obtained with the largest dose (10 S.—N.). No tumor could be made out in any animal 5 weeks after treatment with 10 S.—N. Later on 2 tumors, all told, were manifest in these animals after an incubation period of 3 months, while the average incubation time in the controls was 3—4 weeks. The incubation is also considerably protracted after treatment with 6 S.—N.; more than half of the tumors do not appear until after the 5th week. In the animals treated with 2 S.—N. the tumors appeared on an average one week later than in the control animals.

Table II gives the percentage of animals in which growth of the transplants took place, and the average life-time for the animals that died from their tumors.

The table gives also the number of animals in which the tumors went away again and, in parenthesis, the number of animals wherefrom the average life-time is calculated.

Table II.

	Takes in	Average life-time.
Controls	25 out of 36 — 69.4 %	71.1 days. (24 animals)
	Tumor disappeared in 1.	
2 S.-N.	11 out of 16 — 68.8 %	74.5 days. (10 animals)
	Tumor disappeared in 1.	
6 S.-N.	11 out of 17 — 64.7 %	115.6 days. (11 animals)
10 S.-N.	2 » » 16 — 12.5 »	186.0 days. (2 animals)



Schema II. Carcinoma. Local irradiation shortly after transplantation. Tumors in ¼ natural size 5 weeks after transplantation.

The percentage of growth is not affected by 2 and 6 S.—N., but greatly reduced by 10 S.—N.

The life-time is not definitely influenced by 2 S.—N., but considerably prolonged by 6 and 10 S.—N. In the two animals with growth of transplants after treatment with 10 S.—N., one died with a large tumor 240 days after transplantation; the other mouse died with a tumor, measuring 3×3 mm., 132 days after transplantation. Microscopic examination proved the tumor to be a carcinoma.

Like the preceding series, these experiments show an increase in effect when the dose is increased and, particularly, a marked increase in effect when the dose is increased from 6 to 10 S.—N.

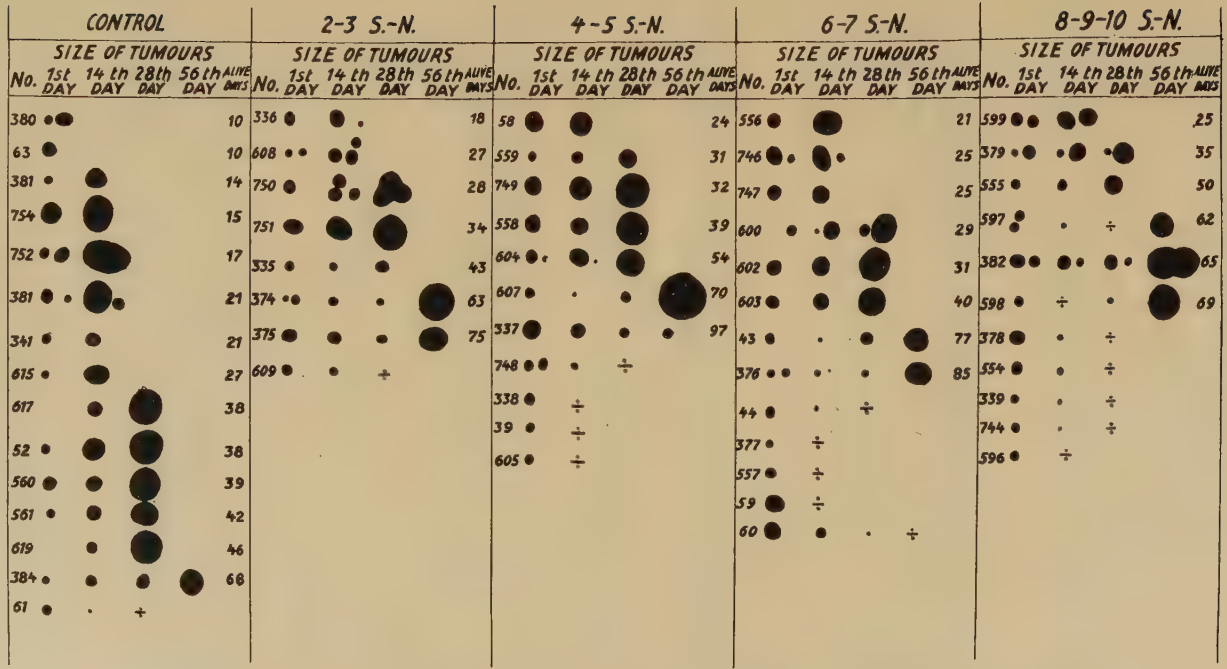
90 animals were used for this series. All the animals that did not die within 5 weeks after the transplantation are included in the calculations, excepting one mouse, treated with 2 S.—N., that escaped from the cage.

The tumors disappeared in 1 control animal and in 1 animal that was treated with 2 S.—N. Autopsy, 5 months later, showed no abnormality in these two animals.

In the cases where the transplants did not grow, the controls and the animals treated with 2 S.—N. lived more than 5 months after transplantation, excepting one control animal that died $3\frac{1}{2}$ months after transplantation. In the 6 animals that were treated with 6 S.—N., and in which the transplants did not grow, 5 lived $6\frac{1}{2}$—8 months after transplantation, while 1 died 5 months after. Out of 14 animals treated with 10 S.—N. in which the transplants failed to grow, 7 were killed 8 months after transplantation; the other 7 had lived, respectively, 7, 6, $5\frac{1}{2}$, 5, $4\frac{1}{2}$, $3\frac{1}{2}$, and 3 months after transplantation. On autopsy none of them showed any indication of tumor.

4. *Experiments with the Sarcoma. Irradiation after Manifestation of Tumor.*

As already mentioned, the sarcoma was less readily influenced by irradiation than was the carcinoma. At first I made some experiments with transplants in the breast; but here the effect of the treatment was very slight, and the animals could not stand the large doses. Difficulties were likewise encountered with the tumors that were implanted in the thigh with a cannula. They appeared so early that it was hard to distinguish between real tumor and tissue reaction following transplantation; their growth was very



Schema III. Sarcoma. Local irradiation after manifestation of tumor. Tumors in $^{1}/_{4}$ natural size.

rapid and malignant, and they reacted but little to irradiation. Injection of tumor-cell suspensions, after grinding with sand or kieselguhr, resulted in a few tumors which developed more slowly. I have made some small series of experiments with such tumors but had to give them up for reasons already mentioned.

This series belongs to my earliest experiments, and they were made at a time when I had not yet decided what doses to employ. So the animals are gathered in groups with gradually increasing doses.

This series, moreover, has the shortcoming that the individual sub-groups do not contain the same number of animals — this was partly due to scarcity of material, partly to indecision about experimental conditions — so there is a source of error in the varying malignancy of the tumors. Still, there were control animals in every one of the smaller series wherefrom the total results is computed.

From Schema III it is evident that in this series, too, an increase in dose gives increase in inhibition of growth, although less pronounced than in the case of the carcinoma.

Table III gives the number of animals in which the tumor disappeared, and the average life-time for the animals that died from their tumors.

Table III.

	Tumor disappeared in	Average life-time with tumor.
Controls	1 out of 15 — 6.7 %	29 days.
2—3 S.-N.	1 » » 8 — 12.5 »	41.1 »
4—5 S.-N.	4 » » 11 — 36.4 »	49.6 »
6—7 S.-N.	5 » » 13 — 38.5 »	41.6 »
8—9—10 S.-N.	5 » » 11 — 45.5 %	51.0 »

The number of animals in which the tumor disappeared shows an upward tendency as the dose increases; it is rather high, though, even after treatment with 4—5 S.—N. The average life-time is greatest after the largest doses, although it is practically just as high after a dose of 4—5 S.—N.

All told, the strongest effect is apparently obtained with the largest dose. The small size of the material, however, and its shortcomings mentioned exclude that any decisive importance be attached to these experiments. But, at any rate, they do not make against the largest doses being the most effective also in the case of the sarcoma.

One thing that may influence the life-time is the fact, that the sarcoma very often produces metastases, which frequently — particularly in these animals of relatively long life-time — become so large that they unquestionably are a contributory cause of death.

This series comprises in all 58 animals. The various doses were distributed as follows: 6 animals got a dose 2 S.—N., 2 got 3 S.—N. 2 got 4, 9 got 5, 6 got 6, 7 got 7, 1 got 8, 2 got 9, and 8 got 10 S.—N.

Out of the 16 animals, in which the tumors disappeared, 14 lived 5 months or more. 1 mouse that was treated with 5 S.—N. died 1½ month after disappearance of tumor, and 1 mouse, treated with 10 S.—N., died after 4½ months. Autopsy revealed no macroscopic abnormality in the last two animals.

5. *Experiments with the Sarcoma. Irradiation Shortly after Transplantation.*

Some of the experiments in this series, which also belong among my earliest experiments, were made with tumors that were transplanted into the breast in the manner described above. Consequently, it was not practicable to employ any dose higher than 6 S.—N. The great majority of animals died after application of 8 S.—N. to the chest. The effect of the higher doses is studied in a supplementing series of experiments with transplantation into the thigh.

In the first series I have used doses of 2, 4, and 6 S.—N. The total results are made up from 5 smaller series, each of which gave the same result.

Schema IV gives the outline of the tumors, reduced to ¼ natural size, on the 13th day after transplantation. The 5 smaller series are kept separate. The size of the tumors is unquestionably decreasing as the dose is increased. (÷) after the tumor indicates the subsequent disappearance of tumor.

Table IV gives the average life-time for the animals that died with tumors, and the number of surviving animals in which the tumor failed to develop to palpable size, or in which a palpable tumor later went away. Each series is listed separately so as to show their conformity.

Table IV.

Exp. No.	Controls	2 S.-N.	4 S.-N.	6 S.-N.
I.	24.7 days	—	32 days 1 living	—
II.	28.3 »	34.7 days 1 living	42.3 days	53 days 1 living
III.	29.7 »	40 days 1 living	43.3 »	48.3 days
IV.	25.3 »	31 days	43.3 »	46.7 » 1 living
V.	27.1 »	30.5 »	38 »	44.4 days
Total	27.1 days 0 living	33.6 days 2 living	40.8 days 1 living	47.5 days 2 living

The transplants grew in all the control animals. 5 of the treated animals survived, 1 or 2 in each group; in 4 of these the tumor developed but disappeared later on. Only 1 animal, treated with 6 S.—N., did never show any sign of tumor.

The life-time is increased steadily with increase of dose. As the various groups do not comprise exactly the same number of animals, I have calculated the average life-time of the controls which correspond exactly to each group of treated animals; as these values vary only from 27.2 to 27.6, the matter may be left out of account.

The total result, then, must be that an increase of dose (up to 6 S.—N.) is associated with an unquestionable steady increase in effect on the sarcoma, too, when it is irradiated shortly after transplantation. Also the smallest dose employed, 2 S.—N., has given a definite inhibition of growth in tumor — and no sign whatever of a more rapid growth.

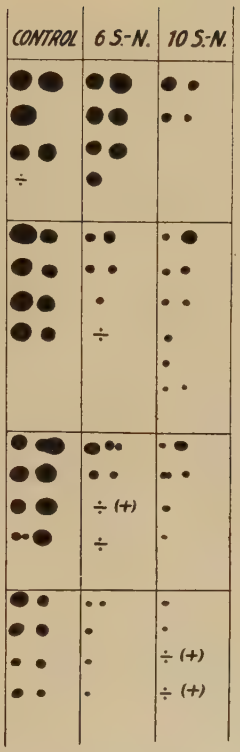
This series was made with 86 animals. Every animal that did not die within 13 days after the transplantation is included in the result, excepting 4 control animals (3 in Exp. No. I and 1 in Exp. No. V) that were killed for transplanting purpose. These 4 animals are listed on Schema IV, but not included in the calculation of average life-time.

The 5 animals, in which tumor disappeared or failed to appear died or were killed 2—4 months after tumor had disappeared. Autopsy revealed no macroscopic abnormality at the site of tumor in any of them.

To supplement this series some sarcomata were transplanted into the thighs, and radiation treatment with doses of 6 and 10 S.—N. was given the 2′ or 3′ day after transplantation. The experiments fall in 4 smaller series.

Schema V gives the outline of the tumors 17 days after transplantation, reduced to $^1/_4$ natural size. The sign -: means absence

Schema IV. Sarcoma. Local irradiation shortly after transplantation. Tumors in 1/4 natural size on the 13th day after transplantation.



Schema V. Sarcoma. Local irradiation shortly after transplantation. Tumors in 1/4 natural size on the 17th day after transplantation.

of tumor, ÷ (+) means a later appearance of tumor. The tumors treated with 10 S.—N. are a trifle smaller than those treated with 6 S.—N.; but the difference is very slight.

Table V gives the number of animals in which the transplants did not grow, and the average life-time for the animals that died. Like in Table IV, the smaller series are listed separately.

Table V.

Exp. No.	Controls	6 S.-N.	10 S.-N.
I.	22 days 1 living	37 days	41.5 days
II.	24 days	38 » 1 living	39.5 »
III.	25.8 »	37.3 days 1 living	47.8 »
IV.	36 »	45 days	55.3 »
Total	27.3 days 1 living	39.6 days 2 living	45.7 days 0 living

The transplants failed to grow in 1 control animal, in 2 animals treated with 6 S.—N., and in no animal that was treated with 10 S.—N. As these figures are so small, and as each group comprises but 16 animals, the margin of accidents is so wide, I think, that one should not attach any great importance to the fact that the transplants grew in all the animals of the last group.

As to the average life-time, it is longest in the animals that were treated with 10 S.—N., a fact that is brought out by each of the smaller series, too. The difference is not very large; still, it certainly is of some significance in view of tumor being remarkably malignant and metastases being present in nearly all the animals, and, besides, it is found in all 4 experiments. That the metastases have been a contributory cause of death is also indicated by the fact that the animals treated with 10 S.—N. generally died with smaller tumors than did the animals treated with 6 S.—N., and, in addition, that the treated animals had smaller tumors than the control animals.

The experiments indicate that in the case of the sarcoma, too, the radiation effect increases as the dose increases up to 10 S.—N.

The series comprises 48 animals. Every animal that did not die within 8 days after transplantation is included in the results. The 3 animals, in which the transplants failed to grow, lived 3—3½ months after transplantation.

C. DISCUSSION OF THE RESULTS

As is evident from the review of previous investigations on irradiation of malignant transplanted tumors in animals, most of the findings seem to indicate that the greatest effect is obtained with the largest doses. Really, KOK & VORLAENDER are the only authors to find that the effect of middle-sized doses is greater than the effect of very large doses. On the other hand, the studies by KOK & VORLAENDER are the most extensive among prior investigations, and they are aimed especially at elucidating the difference in effect of doses of different sizes.

The findings by KOK & VORLAENDER cannot — on basis of their reports — be estimated in the same manner as done in this work, with regard to rate of growth of the tumor, life-time of the animals and number of tumors that have disappeared. For a great many of their animals have died a few days after irradiation — particularly after the large doses — and probably on account of the treatment; and many of their animals were killed at an early period for microscopic examination. In going through their schemas with a view to the macroscopic results, one does not find any proof of a decreasing effect from the largest doses; one rather gets the opposite impression. Besides, the number of animals listed is too small to allow of any definite conclusions from the macroscopic findings. Whether the microscopic changes, whereto the authors attach such great importance, constitute a conclusive evidence, that is harder to say. But, the significance of the histological changes after irradiation is still a question on which opinions differ to such an extent that one has to be very guarded in their interpretation; and it has not been proved that the intensity of such histological changes runs parallel with the therapeutic effect of the Roentgen rays upon the tumor animals.

The easily observable, macroscopic criteria that are used in the present work, the larger material and its greater homogeneity, (since the many Roentgen deaths are avoided and the animals are not killed), and the regular and uniform outcome of the experiments — all these factors warrant, I think, of more sure conclusions.

One must also allow for the possibility that the different ray qualities may differ in effect. Still, I do not find this possibility to be a very likely explanation of the difference in results. For there is only a very slight difference between the ray qualities employed

in the two studies; and there is no base for the assumption of any essential difference in effect of such adjacent ray qualities.

The possibility of the different tumors behaving in different ways does likewise seem less probable to me. The different tumors I have made use of do not furnish any base for such an assumption.

There is a third possibility: that KOK & VORLAENDER have applied the Roentgen rays to such a large part of the mouse that its general health has been affected by the large doses so as to lower its power of resistance against the growth of tumor. Their many Roentgen deaths might be somewhat suggestive of that possibility; but, then it is hard to see how they could get a more favorable effect by treating a large field than by irradiation of a small field.

As to the size of dose, KOK & VORLAENDER emphasize in particular that they have observed the best effect from doses close to or just below the epilation dose for mouse. According to this, the optimal dose in my experiments would be about 5 S.—N.; but there is an increase in effect with doses ranging all the way up to 10 S.—N. The doses given in the two studies likewise agree in their relation to HED for man.

Thus the present experiments indicate that in malignant transplanted tumors of mice the Roentgen treatment with local irradiation gives so much better results as the dose is increased — up to a dose that is twice the epilation dose, and which causes serious injury to normal tissues surrounding the tumor.

It is not probable that a dose of such size is capable of lowering the resistance of the organism to the tumor by lowering the resistance of the normal tissues in the treated area.

In addition, these experiments imply the conclusion that: there is no definite curative or lethal dose, common to all animals — not even when the same tumor strain is employed throughout. The outcome is gradually better as the dose is increased.

IV. ON THE EFFECT OF UNIVERSAL RADIATION TREATMENT UPON TRANSPLANTED TUMORS IN ANIMALS

A. PREVIOUS INVESTIGATIONS

It has been known for a long time that animals do not tolerate large doses of universal Roentgen radiation. They die shortly after such treatment, and on autopsy they show various pathological changes, particularly in the blood and the hemopoietic organs (FABRICIUS-MØLLER, 37). From their experiments WARREN & WHIPPLE (175) feel justified in concluding that the death is chiefly due to epithelial destruction of crypts and villi in the small intestine and following intoxication.

I have myself seen numerous deaths in mice after universal irradiation and after local treatment with excessive doses and too large a field. Very frequently the animals had diarrhea; and on autopsy I often found hemorrhages of the intestines, the subcutaneous tissue and other parts — even of the brain, occasionally. With the radiation factors I have been using, the animals usually die from universal treatment with 3½ S.—N. through 5 mm. Al, and, as a rule, within 15 days.

Many investigators have given universal treatment to animals, mostly mice, with malignant — mostly transplanted — tumors. Most of the experiments are dealing with irradiation of the animals before transplantation to study the influence of Roentgen treatment on immunity towards neoplasms.

MURPHY & MORTON (112) have shown that spontaneous immunity as well as the immunity produced by injection of homologous living tissue may be broken down by repeated universal treatment with fairly small doses of soft Roentgen rays. They think that the reason for this is: that the lymphocytes are destroyed by the treatment. After a while, when regeneration of lymphocytes has taken place, the animal becomes immune once more.

MURPHY & TAYLOR (116) showed that the same holds good for animals that are immunized artificially against a transplant-

able carcinoma and, on previous inoculation, have proved to be immune to the tumor strain in question.

Murphy, Hussey, Nakahara & Sturm (110) found that mice, which had been sensitized by injection of rat blood, were immune to cancer transplants when rat blood was injected at the time of transplantation. This immunity would break down on repeated exposures to Roentgen rays.

Murphy, Nakahara & Sturm (114) applied a small dose of X-rays to the back of some mice. When such mice were inoculated with a carcinoma immediately after the treatment, they showed some increase in susceptibility; those inoculated 7 days after treatment showed a decrease in susceptibility.

Nakahara & Murphy (121) found the refractory state to last from the 3rd to the 7th day after treatment. In the susceptible state there was no lymphocytic infiltration at the site of inoculation, while it was very marked in the refractory state.

The same authors (122) found an increase in lymphocytes lasting from the 2nd to the 10th day after treatment, when they treated the mice with a definite, very small, dose of soft rays from a special thin-walled tube. In the same period there was an increase in the number of mitoses in the lymphoid tissue. — Animals that were treated in this way showed an increased resistance to cancer transplants, beginning on 3′ day, but most pronounced on 10′ day.

Taylor, Witherbee & Murphy (168) demonstrated, in various animals, a marked fall in the number of lymphocytes after universal irradiation with fairly large doses. Minimum was reached in 48 hours.

Thomas, Taylor & Witherbee (170) treated 9 rabbits with small doses of soft Roentgen rays. Blood count showed the number of lymphocytes to have increased considerably in 5, and definitely in 2 of the animals. 2 animals that were treated with harder rays, filtered through 5 mm. Al, showed no increase of number. Nakahara (120) examined the lymph glands of these animals and found no destruction but an immediate stimulation, most pronounced 2—4 days after treatment and lasting 2 weeks.

Murphy & Morton (113) made 3 series of experiments on mice with spontaneous cancer, wherein the tumor was removed and a small autograft implanted in inguen: in the 1′ series after universal treatment of the animals with a small stimulating dose of Roentgen rays, while the tumor was removed; in the 2′ series with no treatment of animal or tumor; in the 3′ series after treatment of tumor *in vitro* with the same dose. The 2′ and 3′ series showed no diffe

rence in frequency of growth; the first series showed a considerable degree of immunity and a smaller number of recurrences of the removed tumor.

In this series of splendid investigations at the Rockefeller Institute these authors have thus demonstrated that universal irradiation with small doses of soft Roentgen rays will increase the resistance to subsequent cancer transplants. The same holds true for autografting; and this treatment also lowers the percentage of recurrency after removal of a spontaneous tumor. They have also shown, on the other hand, that larger doses lower the resistance. They are of the opinion that the variation in resistance runs parallel with the variation in number of lymphocytes, as large doses destroy the lymphocytes while small doses stimulate the activity of the lymphoid organs and increase the number of lymphocytes.

Their experiments have been confirmed in part by others; but, the rôle of the lymphocytes is still a matter of dispute among various investigators.

Sittenfield (164) made his experiments with Flexner-Jobling's rat sarcoma and found that naturally immune rats, whose immunity was controlled through repeated implantations, did not loose their immunity on repeated universal irradiations, which lowered the number of lymphocytes. The rays he used were rather hard.

Mottram & Russ (108) were able to destroy the natural immunity of rats against Jensen's rat sarcoma by treatment with large doses, which produced a decrease in the number of lymphocytes. Many of the subsequent takes subsided as the lymphocytes began to increase in number.

Russ, Chambers, Scott & Mottram (151) found that quite small daily doses of X-rays, characterized as medium-soft, gave immunity without any demonstrable increase in lymphocytes.

Prime (139) did not find any loss of natural or artificial immunity after treatment with X-ray doses that lowered the number of lymphocytes. He has made large series of experiments on rats with different tumors. His total dose is 10 H, unfiltered, of rather soft rays within 5 days.

Caspari (24) was able to break down the natural immunity of mice to Ehrlich's carcinoma by treatment with strong universal Roentgen doses. A very small single dosis resulted several weeks later in marked leucocytosis, at which time the resistance was found to be increased.

Universal irradiation of animals after development of the transplanted tumor has not been carried out to any great extent.

In 1909, Meisen Westergaard (102) exposed mice with Jensen's carcinoma to universal irradiation with a daily dose of 1¼ S.—N. for 9 days, given shortly after transplantation, and found as result: prolongation of incubation time, inhibition of tumor growth, and prolonged life-time of the animals. A single dose of this size showed the same effect, although less pronounced and decreasing on increase of the interval between transplantation and irradiation.

Levy-Dorn (94) gave universal treatment to carcinoma- and sarcoma-mice and found that they could not stand as large doses as were required for making tumor disappear.

Russ, Chambers & Scott (150) treated a series of rats by giving them small universal daily doses of 12 sec. (spark gap of 4.5 cm. between spheres of 5 cm. in diameter, target distance 20 cm., unfiltered). The treatment started before transplantation and was continued for 4—8 weeks. The transplants grew more slowly than in the control animals, and more of them subsided than usual. The tumor itself was rather disposed to spontaneous disappearance. When tumor was screened, universal irradiation, begun after the appearance of tumor, 12 sec. a day for 1 month resulted in some delay of growth and disappearance of more tumors than in the controls. When tumor was not screened, universal irradiation, begun after the appearance of tumor, 5 min. a day for 1 week had considerable effect on the tumor, but the animals died from the treatment. On treatment 1 min. a day for 1 month, there was some effect on the tumor, but the animals lost weight. On 12 sec. a day there was less effect on tumor (also less than on the screened tumor) but the animals kept their weight. On 2 sec. a day for 4 weeks there was no definite effect. Previously these authors had shown that on daily treatment for 1 min. normal animals gained weight more slowly than did the non-treated control animals; on daily treatment for 2—12 sec. the normal animals gained weight faster than the controls. The authors have also found that treatment of tumor *in vitro* with very small doses causes a more rapid growth of tumor on subsequent implantation in non-treated animals. — Their conclusion is that one must avoid the small doses in local treatment of the tumor and the large doses in universal treatment on the other hand, there may possibly be indications for universal treatment with small repeated doses.

Valken (171), too, was able to increase the resistance in mice against carcinoma and sarcoma by universal Roentgen treatment with small repeated doses on irradiation before as well as after transplantation.

In their first experiment KOK & VORLAENDER (75) gave local treatment to normal mice and found some histological changes, not only in the exposed area but also in the non-treated part of the skin. The latter changes were of the same character as those in the treated area, differing only in degree. They are interpreted as resulting from the general effect of the treatment. Next (76, 73, 74, 173) they gave universal treatment in various doses to mice with Ehrlich's carcinoma, partly after the appearance of tumor, partly shortly after transplantation, and partly before transplantation. In every instance they found a considerable effect upon the tumor, just as good as that they have obtained on local treatment. In the animals that were treated after the appearance of tumor, the effect from 25 e was only slight, but treatment with 50 e — *i. e.*, 20 per cent of the epilation dose — was just as effective as local treatment with a dose four times as large. All these animals, however, died within a short time after irradiation. — In animals that were treated shortly after transplantation the best effect was obtained with 10 per cent of the epilation dose. On irradiation before transplantation the most effective dose was 10 e — *i. e.*, 4 per cent of the epilation dose — while 25 and 50 e were less effective. Universal irradiation was also effective — though to a less degree — when the implantation area was screened. From these and other experiments the authors conclude that the size of dose is of less significance than the irradiated volume, and that the effect of the rays is mainly an indirect one. — In difference from English and American investigators, KOK & VORLAENDER used very hard rays, and they gave only single doses.

In 1922, CASPARI (24) reported that he had found no definite effect from small universal irradiations in manifest tumors because, he thinks, the tumors grew so rapidly that the animals died before the number of lymphocytes was in the positive phase. In 1924, CASPARI (26) stated that, like KOK & VORLAENDER, he had found just as good or — as he says himself — just as poor effect from universal irradiation as from local treatment. — He has also seen spontaneous tumors of mice diminish after universal irradiation, but never disappear altogether.

Most of these experiments indicate that small universal Roentgen radiations, preferably repeated for some time, may have a favorable therapeutic effect in transplanted tumors of mice and rats, while large doses do harm. It is true that a large dosis may have a favorable effect as to the tumor, but the animal falls off, and local application of the same dose must be assumed to have a better effect. KOK & VORLAENDER differ somewhat from most other authors in using

such large doses that one would expect a pernicious effect instead of a beneficial one or, at any rate, not as good effect as obtained with local treatment.

B. OWN INVESTIGATIONS

1. Arrangement of Experiments.

The general conditions regarding material, technique and arrangement of experiments are the same as those mentioned in the preceding chapter.

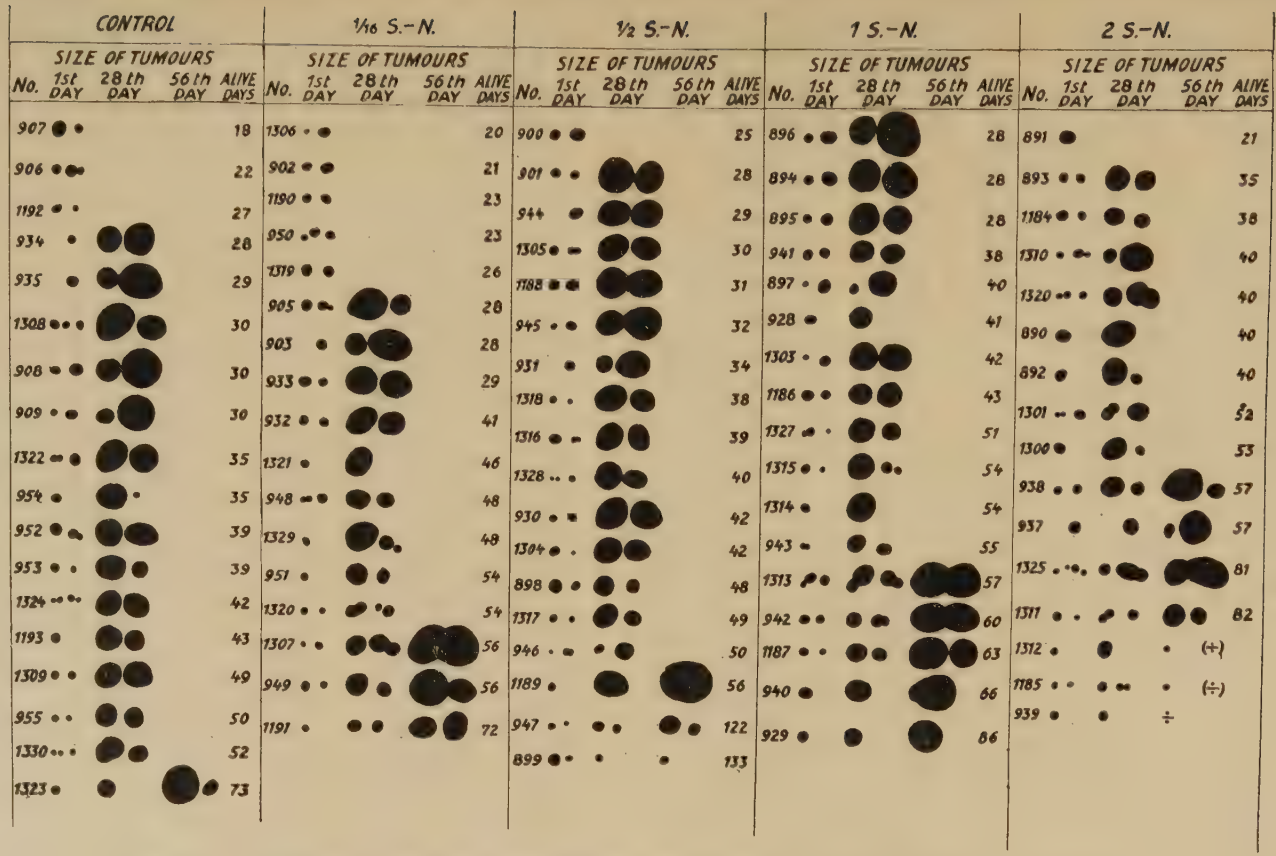
For universal irradiation the non-tied animals were placed in a box with 4 small compartments, each of which was just large enough to hold one mouse. Usually they were treated 4 at a time. The box was made of wood, cardboard and adhesive tape; it had no metallic part. The animals were supplied with fresh air through a narrow slit in the head-end of each compartment. The box was placed straight under the tube so that the distance from focus to the back of the animals was 23 cm.

On these experimental conditions the animals die, as a rule, from $3^1/_2$ S.—N. For maximal dose I have used 2 S.—N. The great majority of mice survive a dose of this size. I have treated the animals with single exposures only.

2. Experiments with the Carcinoma. Irradiation after Manifestation of Tumor.

This series comprises 5 groups of animals, 1 control group and 4 groups of animals treated with $^1/_{16}$, $^1/_2$, 1, and 2 S.—N. The series is made up of several smaller series wherein the groups have the same number of equally large animals with tumors of the same size. As only very few animals died so early that they had to be left out of the account, the individual groups correspond closely to each other.

Schema VI shows the outlines of the tumors, reduced to $^1/_4$ natural size, at the beginning of the experiments and at the end of 4 and 8 weeks. The various groups do not differ much in size of tumor. Still, they show a tendency of tumor to slow down in growth as the dose is increased; and there is a definite difference in size of tumor between the controls and the animals treated with 2 S.—N.



Schema VI. Carcinoma. Universal irradiation after manifestation of tumor. Tumors in 1/4 natural size.

Table VI gives the number of animals in which the tumor has disappeared and the average life-time of the animals that died with tumor.

Table VI.

	Tumor disappeared in	Average life-time
Controls	0 out of 18	37.3 days
1/16 S.-N.	0 » » 17	39.6 »
1/2 S.-N.	0 » » 18	48.2 »
1 S.-N.	0 » » 17	49.1 »
2 S.-N.	3 » » 16	48.9 »

The tumors have disappeared in 3 animals, all told; and these 3 animals all belong to the group that was treated with 2 S.—N. Compared with the findings after local treatment, these tumors were unusually slow in disappearing.

The 3 last groups show a slight rise in the average life-time. This rise does nowhere exceed the increase obtained with local application of 2 S.—N. in previous experiments with the same tumor. The comparatively long life-time of the group that was treated with 1/2 S.—N. is chiefly due to 2 animals which lived unusually long. The increase of life-time is allotted more evenly in the other groups.

Universal irradiation has thus been demonstrated to have some beneficial effect, but — with the doses employed — no more than could be expected from local treatment with the same doses, and far from as much as may be obtained with local application of larger doses.

The results cover 86 animals. 5 animals were left out of the final results because they died within 10 days after irradiation; they were: No. 926, 927, and 936, treated with 2 S.—N.; No. 1302, treated with 1 S.—N.; and No. 904, treated with 1/16 S.—N.

The 3 animals, in which the tumors disappeared, died 2, 4, and 4 1/2 months later. Autopsy revealed no sign of cancer in them.

3. *Experiments with the Carcinoma. Irradiation Shortly after Transplantation.*

These experiments are in keeping with the corresponding experiments on local irradiation. They were carried out concurrent with the latter, in the same 3 smaller series, and with the same control animals.

The doses employed are 1/4, 1/2, 1, and 2 S.—N. The animals were treated 2—3 days after transplantation.

Schema VII gives the outline of the tumors, reduced to 1/4 natural size, 5 weeks after transplantation. The sign ÷ indicates the absence of a tumor at that time; ÷ (+) means the later appearance of a tumor; + signifies that the animal had died with a macroscopic tumor which, on microscopic examination, proved to be identical with the one implanted; »died» denotes that the animal died without any demonstrable tumor. (÷) after tumor signifies its subsequent disappearance.

In the first series the tumors come near being a little smaller in the treated animals than in the controls, but the reverse is rather the rule in the other series. All told there is not a greater difference than what may be due to individual variations. Nor do the treated groups differ to any distinct degree. The incubation time was not affected by the irradiation as was the case in the experiments with local irradiation.

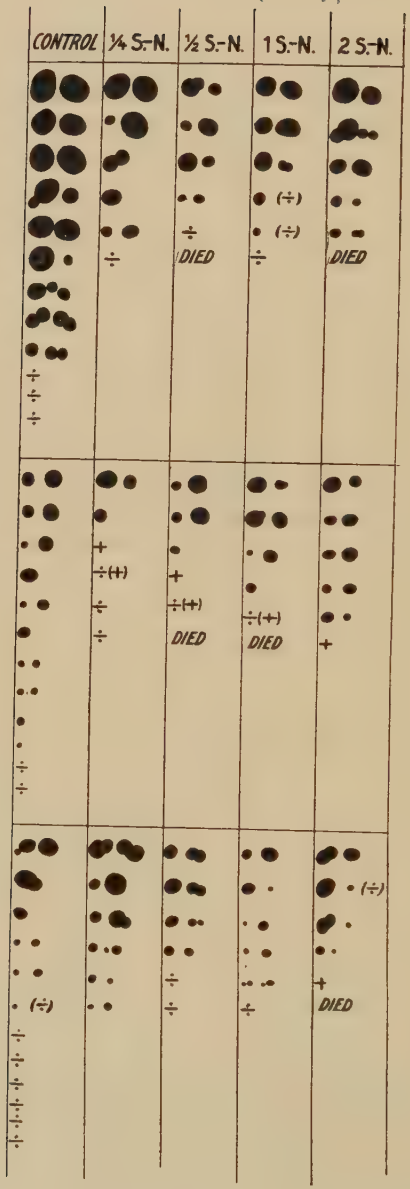
Table VII gives the percentage of takes and the average life-time of the animals that died with tumors.

The table also gives the number of tumors which subsequently disappeared and, in parenthesis, the number of animals from which the average life-time is calculated.

Table VII.

	Growth in	Average life-time
Controls	25 out of 36 — 69.4 % 1 disappeared	71.1 days (24 animals)
1/4 S.-N.	15 out of 18 — 83.3 »	74.4 » (14 »)
1/2 S.-N.	13 » » 16 — 81.3 »	79.8 » (12 »)
1 S.-N.	15 » » 17 — 88.2 » 2 disappeared	79.3 » (13 »)
2 S.-N.	16 out of 16 —100 » 1 disappeared	71.2 » (13 »)

The percentage of takes is found to be higher in the treated groups than it is in the controls, while it was found to be lower in the corresponding groups that were treated with local application (p. 40); and, on universal irradiation, there is a tendency to increase in percentage of takes as the dose is increased to 2 S.—N., in which group the transplants took in all the animals. Even though there are but 16—18 animals in each of the treated groups and twice as many control animals, the increase in percentage of takes can hardly be the outcome of chance. Addition of all the animals treated with universal application gives a total of 59 takes in 67 animals — that is 88 % — numbers, where the individual diffe-



Schema VII. Carcinoma. Universal irradiation shortly after transplantation. Tumors in 1/4 natura size 5 weeks after transplantation.

rences can hardly assert themselves. There is also a marked difference in percentage of takes between treated and non-treated animals, even when the animals in which tumor disappeared are left out. This difference can hardly be due to any other factor than this: universal irradiation has lowered the resistance of the animals to the tumor. — In these groups comparatively many tumors have disappeared later on, a fact that also is observed by MOTTRAM & RUSS (108), and which presumably comes from the animals regaining their resistance when the radiation effect has passed away.

As to average life-time, none of the groups shows any definite effect from the treatment.

The carcinoma-mice thus fail to show any beneficial effect from universal irradiation with the given doses shortly after transplantation. This treatment, on the other hand, is found to lower the resistance of the animals to implanted carcinoma.

108 animals were used in this series. All the animals that did not die within 5 weeks after transplantation, and in which the tumor did not disappear later on, are included in the calculation of the average life-time. 1 mouse that was treated with 1 S.—N. got a tumor, which disappeared later on. 2 months later, a tumor developed at vulva. The animal lived considerably longer than the other animals, and, when it died, it had a large tumor, that had not developed like the other transplants but was extending all around vulva. Microscopic examination proved this tumor to be a cheratinizing squamous cell carcinoma. I have therefore taken it for a spontaneous tumor; and I have omitted the animal from the calculation of average life-time.

All the animals in which the transplants did not take, or in which tumor subsequently disappeared, have lived at least 5 months after transplantation, with the exception of 1 control animal that died $3^1/_2$ months after transplantation.

4. Experiments with the Sarcoma. Irradiation Shortly after Transplantation.

On account of the before-mentioned difficulties in irradiation of the sarcoma, universal treatment has been given only 2—3 days after transplantation, and not to animals with manifest tumors.

The experiments were made with transplants in the breast, and concurrent with the corresponding experiments on local irradiation (Chap. III, 5; pp. 45—47). The same control animals are used for both series, so the results are truly comparable. This series comprises 5 smaller series.

The doses employed are $^1/_{16}$, $^1/_6$, $^1/_2$, 1, and 2 S.—N.

Schema VIII shows the outlines of the tumors (reduced to $^1/_4$ natural size) 13 days after transplantation. No difference can be made out between the various groups of this series, whereas the corresponding experiments with local irradiation (as evident from Schema IV) showed considerable effect on the tumors.

Table VIII gives the average life-time of the animals that died with tumors, and the number of animals in which tumor disappeared. The transplants grew in every animal. The individual series are listed separately to illustrate how they agree.

Table VIII.

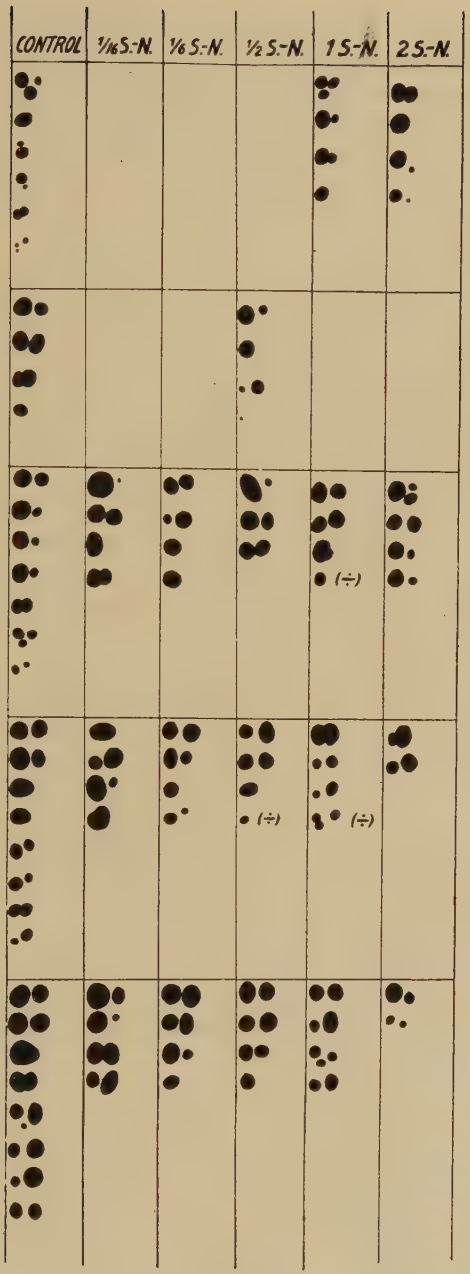
Exp. No.	Controls	$^1/_{16}$ S.-N.	$^1/_6$ S.-N.	$^1/_2$ S.-N.	1 S.-N.	2 S.-N.
I.	24.2 days				24.5 days	24.3 days
II.	23.3 »			26.3 days		
III.	29.7 »	25.0 days	23.3 days	27.8 »	28.0 » 1 living	21.5 »
IV.	25.3 »	27.8 »	29.3 »	22.0 » 1 living	29.7 days 1 living	25.5 »
V.	27.1 »	23.3 »	23.8 »	34.5 days	22.8 days	29.0 »
Total	27.1 days 0 living	25.4 days 0 living	25.4 days 0 living	28.0 days 1 living	25.9 days 2 living	24.3 days 0 living

The various groups show no greater difference in average life time than what may readily arise from individual peculiarities. As the individual groups do not quite correspond to each other, I have calculated the control groups which precisely correspond to each group of treated animals; and as the average values only vary from 26.7 to 27.6, I have felt justified in leaving that out of consideration.

The only suggestion of any beneficial effect is the fact that tumor disappeared in 2 animals that were treated with 1 S.—N. and in 1 animal treated with $^1/_2$ S.—N. It did not disappear in any of the 33 control animals. This tumor is very malignant, and its disappearance is such a rare occurrence that the treatment may conceivably have caused the tumor to disappear in these 3 instances. However, many more experiments are required for a definite view in this respect.

No definite — favorable or noxious — effect of the treatment could be demonstrated in this series. The corresponding experiments with local irradiation show a definite effect.

This series comprises 110 animals, all of which are entered on Schema VIII. 3 of the control animals in Exp. I and 1 in Exp. V



Schema VIII. Sarcoma. Universal irradiation shortly after transplantation. Tumors in 1/4 natural size on the 13th day after transplantation.

were killed for further transplantation, wherefore they do not enter in the calculation of the average life-time.

The 3 surviving animals lived respectively 2½, 3, and 6 months after the tumor had disappeared to impalpability. Autopsy revealed no macroscopic abnormality in area of transplantation and no evidence of metastases.

C. DISCUSSION OF THE RESULTS

It is interesting to make a comparison between the animals that were treated locally with 2 S.—N. and those exposed to universal irradiation with the same dosis shortly after transplantation. In these series the experiments with local and with universal treatment were made at the same time and with the same control, wherefore their results are comparable. The dosis applied to the tumor cannot vary much in the two series. The only difference in experimental conditions is this, that universal treatment takes in a larger field of irradiation. This will cause an increase of dosis, if anything, so that the transplants in the animals treated with universal irradiation most likely got a slightly larger dose than did those exposed to local application only. There can hardly have been any appreciable influence on the results from the fact that the animals in one series were stretched out and in the other were left loose in small compartments that did not leave much room for moving around.

As illustrated, the tumors are largest in the animals treated with universal irradiation. This is particularly conspicuous in the case of the carcinoma, but it also applies to the sarcoma. As to the sarcoma, the percentage of takes is 100 in either series, but tumor disappeared in 2 animals that were treated with local application and in no animal exposed to universal irradiation with 2 S.—N. In the case of the carcinoma the percentage of takes is 100 on universal treatment and only 68.8 on local treatment. The average life-time of the sarcoma mice after universal irradiation is 24.3 days (control: 26.7 days), after local treatment it is 32.6 days (control: 27.6 days). The average life-time of the carcinoma mice after universal irradiation is 71.2 days, after local treatment it is 74.5 days; the numbers of survivals are, respectively, 1 and 6.

So universal irradiation did away with the favorable effect that would have resulted from local treatment with the same dosis. This also demonstrates the injurious effect of universal irradiation with large doses.

I have not made any experiment with irradiation before transplantation, wherefore my experiments are not directly comparable to the irradiation experiments by MURPHY and co-workers and by other investigators. It is not certain that irradiation acts in the same manner whether it is given before or after transplantation. According to CASPARI (25), any treatment, which — like universal X-ray treatment — causes cell destruction in the organism, produces an increase in the resistance to subsequent cancer transplantation. But, these methods have so far proved impracticable in the therapy.

Other experiments with irradiation after transplantation show — like mine — that the resistance of the organism to the transplanted tumor is lowered after irradiation with larger doses. There is nothing in my experiments, on the other hand, to indicate that the resistance is increased after small doses. Such an effect has apparently been demonstrated to a certainty by RUSS, CHAMBERS & SCOTT and by MURPHY & MORTON. The absence of this effect in my experiments may be due to the fact, that the small doses I have employed have not been of the proper size to produce this effect, or that I have only given single doses, or that the ray quality has been different. The American investigators in particular have used a very soft ray quality, and their experiments indicate that the effect is so much the better as the rays are softer. The ray quality I have been working with has been rather hard. It is very difficult to compare the doses. At any rate, the active dose appears to be very small and very limited.

MEISEN WESTERGAARD'S results are explainable as the outcome of the local effect alone and are not in conflict with my findings.

KOK & VORLAENDER'S results, on the other hand, do not agree with mine. The ray quality is pretty near the same in both investigations, and the proportion between the doses employed and the epilation dose for mouse warrants a comparison between the doses. They claim to have demonstrated an excellent effect from one single exposure to universal radiation with 20 % of the epilation dose in the case of manifest tumors, and with 10 % of this dose on irradiation shortly after transplantation. I have not been able to confirm this statement. The mentioned doses correspond to about 1 S.—N. and ½ S.—N. in my experiments. For reasons pertaining to the experimental conditions, which I have already discussed, I think that the more trusty conclusions as to the therapeutic effect may be drawn from my experiments.

I have not been able to find any account of CASPARI'S aggre-

gate material. From his own statement the result seems to b somewhat uncertain, but why his findings should possibly differ fro mine, that I cannot say.

These experiments, I think, demonstrate that single universal irra diations of mice with doses of 1/80—1/3 epilation dose after implan tation of malignant tumors do not produce any favorable therapeuti effect, which in any way approaches the effect that can be obtained b local treatment with larger doses — hardly more than what may b obtained on local application of the same doses. Under certain cir cumstances, on the other hand, universal irradiation may even hav a noxious effect.

It is not likely that the therapeutic effect of X-ray treatment to an great extent is due to the general effect on the organism; more likel it is a question of local effect.

V. ON THE EFFECTS OF THE SAME TOTAL DOSIS DISTRIBUTED OVER DIFFERENT TIMES

A. PREVIOUS INVESTIGATIONS ON NORMAL TISSUES AND CELLS

The studies by KRÖNIG & FRIEDRICH (82) of radiation effect on human skin have largely been instrumental in establishing the claim that radiation treatment of malignant tumors should be carried out with total doses in the shortest possible time. They obtained less effect with the same radium or Roentgen dosis when it was distributed as several smaller doses in several days than when given as a massive dosis. Moreover, they were able to demonstrate that the skin reaction is weaker when the ray intensity is lowered. There was a conspicuous difference when the intensity was reduced to 1: 8, and the radiation time, of course, was 8 times as long.

These findings were confirmed by SCHWARTZ (158) who, in Roentgen treatment, could even demonstrate a difference in skin reaction with an intensity difference of 1: 4. And he always found a stronger reaction from 1 HED × 1 than from ½ HED × 2 at intervals of 24, 6, and 2 hours.

Most radiologists, I think, have had the opportunity of confirming these findings. On distribution of the dosis over a longer period, the skin reaction is weaker than when the same dosis is given in short time.

One cannot, however — like KRÖNIG & FRIEDRICH — claim that when the massive dosis has the strongest effect on the skin, it will also be the best method of application in treatment of malignant tumors. This point depends particularly on how the skin and other normal tissues react to the distribution of dosis in comparison with the reaction of tumor to the same treatment. If the normal tissues and the tumor react alike, it really does not matter how the dosis is given when no other factors are to be taken into account. If, on the other hand, they differ in reaction, there is a possibility of obtaining a relatively greater effect on tumor or on the skin. If the skin recovers its resistance more slowly than does

the tumor, the best therapeutic effect is obtained by a massiv dosis. If, on the other hand, the skin recovers its resistance mor rapidly than the tumor, protracted treatment will give the bes therapeutic effect.

Also another human object, the ovaries, shows less effect whe the dosis is distributed over longer time, as it then takes a large dosis to produce amennorrhea. WINTZ (180) found this »castratio dosis» to be 34 % HED when given in one application, and 50—52 % when distributed over 8 days.

The skin of various animals reacts like human skin on irradia tion. This has been demonstrated by NATHER & SCHINZ (123 with Roentgen treatment of mice, by SAMSSONOW (154) with X rays on rats, by HUDSON (64) with radium on rats, and by REGAU & FERROUX (144) with X-rays on rabbits.

Numerous other investigators have demonstrated the sam conditions of reaction in other animal objects.

In X-radiation on ascaris eggs, HOLTHUSEN (62) obtained les effect with less intensity; he also found the effect of 4 small dose in 90 minutes to be less than that of a dose 4 times as large in minutes and of the same intensity. — When the eggs were kep in an oxygen-free atmosphere during the treatment, whereby thei development was temporarily suspended, there was no differenc in effect with different time distributions or intensities of the sam dose.

MARKOVITS (99) determined the lethal dosis for *Paramecium caudatum* in a certain period of life as 8—10 hours' radiation wit a certain mesothorium preparation. 60 minutes' radiation daily fo 2 weeks, *i. e.* 14 hours altogether, had no effect at all; nor was 9 minutes' radiation daily for 2 weeks — 21 hours all told — enoug to kill the animalcules.

From X-radiation on growing bones in rabbit and cats, HOFF MANN (55) found less effect when the dosis was distributed over week. When it was distributed over 3 days only, there was n difference from the massive dosis.

WARREN & WHIPPLE (175) determined the lethal dosis in X radiation on the abdomen of various animals; they found it to be th same whether it was given in one application or distributed ove the first 4—6 days. When distributed over longer time, the dos could be increased.

LAROQUETTE (89) found less effect with divided doses in X radiation on larvæ of *Rana fusca*.

In a very pretty experiment, ANCEL & VINTEMBERGER (4) hav first treated hen's eggs with X-rays before incubation, and foun

the same effect whether the whole dosis was given at one time or divided in 15 small doses over 8 hours. They then treated 3 groups of eggs with 15 H. before incubation and after incubation for 2 and 9 hours, respectively. A 4′ group was treated with 15 × 1 H between 2′ and 9′ hour. The eggs were examined after further incubation for 3 hours. The 4′ group was least affected. The same experiment with a total dosis of 25 H was still more cogent in its results. Repetition of the experiments gave exactly the same outcome.

In X-radiation, CROWTHER (29) found less effect on the protozoon *Colpidium colpoda* and HANCE & CLARK (50) found less effect on *Paramecia* when the dosis was divided.

Similar conditions have been demonstrated in plants.

GUILLEMINOT (46) applied radiation to dried pumpkin seeds and found a complete cumulation of the divided doses; he showed, too, that the radiation effect was the same whether the seeds were planted at once or not until as much as 2 years later.

Similar results were obtained by JÜNGLING (69) with dried horse-beans. Sprouting beans, on the other hand, showed less effect when the dosis was divided in 4 equal parts at intervals of 3 hours. When the sprouting beans were placed on ice, the radiation effect was the same as in the dried beans.

MATONI (101) has likewise worked with sprouting beans. He found the effect to be less when the ray intensity was less, though the dosis was the same in other respects. The proportion 1: 4 gave a distinct difference in effect.

In Denmark, ARNTZEN & KREBS (11) studied this effect on sprouting peas. They found a decrease of effect when the dosis was divided in 2—4 parts with intervals of 24—4 hours. When the dosis was distributed in 2—3 parts over 24 hours, the difference in the inhibition of growth was observable also after planting, but not so when distributed in 4 parts over 12 hours.

And, finally, SUZANNE ANCEL (5, 6) has demonstrated that the effect of X-radiation on sprouting lentils is lowered by dividing the dosis as well as by decreasing the intensity.

This question is also studied on tissue cultures, where CANTI & SPEAR (23) found less effect from radium treatment with less intensity than one would expect according to physical calculations.

There is a most beautiful conformity in all the mentioned experiments by a great number of investigators on numerous objects. Cells in a latent state show complete cumulation of several radiations, while cells in active state show incomplete cumulation. Even a decrease of ray intensity with consequent prolongation of radiation time gives a decreased effect.

Some other experiments indicate, however, that this is not th case in all objects and under all circumstances.

As early as 1913, LAZARUS-BARLOW (92) made the interestin experiment of placing radium tubes of different strength in rectur of rats for different lengths of time, so that each rat got the sam dosis (measured ionometrically). The inner half of the tube wa acting on the cylindrical epithelium of the rectal mucosa, the oute half on the squamous epithelium of the root of the tail. The great est effect on the cylindrical epithelium was obtained with a sma dosis over long time; on the squamous epithelium the reverse wa the case.

REDFIELD & BRIGHT (141) studied the radiation effect wit radium-emanation capillaries without filter on eggs of *Nereis limbata* When the egg is fertilized, it is surrounded by a fertilizatio membrane, which gets thicker if the egg is irradiated beforehand The increase in thickness of this membrane depends on the size o the dosis. With the same product of intensity and time, the great est effect was obtained with a small intensity over a long time and less effect with a great intensity over a short time. The sam difference in effect was observed in the delayed development o the eggs.

RADOS & SCHINZ (140) have tried to determine the X-ray dosi which produces keratitis in rabbits, and found the massive dosi to be ca. 1,700 % HED. On repeated radiations at intervals o 2—3 days, keratitis was produced by 1,000 %; with longer inte vals it took a larger dosis than on massive treatment. The experi ments are not convincing; the records comprise but 2 animals tha are treated at short intervals, and there are very large individua differences in the results from irradiation with single doses.

In sprouting beans GLOCKER, ROTHACKER & SCHÖNLEBE (44) found no dependence of effect on intensity with the proportio 1: 10. But, they have made only 2 experiments with 5 beans i each; and in their later determinations of the depth intensity b measuring with beans they found some deviating results whic might indicate that there nevertheless was a dependence on in tensity.

RUSS & SCOTT (152) introduced radon capillaries in the live of rats and in Jensen's rat sarcoma. The effect on the liver tissu was found to be almost independent of the intensity; only when th difference of intensity was very marked — the radiation time had t be extended from 2 hours to 30 days — did the smaller intensit have relatively smaller effect. The results on the sarcoma wer more difficult to define, but there did not seem to be any princip

difference. The authors estimate the radiation effect by the extent of the necrotic zone around the radon capillaries.

Like HOLTHUSEN, ZUPPINGER (184) studied the effect of X-radiation on ascaris eggs. Contrary to H., he obtained the strongest effect with a weak intensity over a long time; and fractional radiation distributed over 10 days had about twice the effect of the same total dosis in one application. He thinks himself he has avoided the source of error implied in the varying radiosensitivity of the eggs — a very important matter in experiments of this kind, where the irradiation is greatly protracted.

The most significant and best known experiments in this respect, however, are the radiation experiments on testes by REGAUD and by SCHINZ & SLOTOPOLSKY. On basis of their findings and from the view that the cells are most radiosensitive in the mitotic period, wherefore one must try to hit as many as possible while they are dividing, many radiologists look upon the protracted radiation as more effective than the massive.

Since BOHN (19) and PERTHES (132, 131) published their investigations, in 1903—04, it has been known, that irradiation acts on the chromatin of the nucleus and results in an abnormal course of the mitosis. These studies and subsequent investigations by BERGONIÉR & TRIBONDEAU (17, 18), REGAUD & BLANC (143), and KRAUSE & ZIEGLER (77) have shown that particularly the tissues which are actively proliferating, are most readily affected by radiation. This was thought to be due to a lowered resistance of the cells in the mitotic state. Investigations by MOTTRAM (107), RICHARDS (146), GRASNICK (45), HOLTHUSEN (60) ALBERTI & POLITZER (1), STRANGEWAYS & HOPWOOD (166), *et al.* support the view that cell division is associated with periods in which the cells are particularly sensitive to radiation, perhaps also to other injurious agencies. From this view one would expect the better effect from protracted radiations, as such would hit most cells in the sensitive period.

In his experiments, REGAUD (142) aims to show that such is the case. He has worked with testes of rams that are especially sensitive objects for the effects of radium and Roentgen rays at the time of spermiogenesis. Besides, they present several points of resemblance to malignant tumors in the continual changing of resting cells and dividing cells and in the varying radiosensitivity of the cells that are present at the same time. In the testes REGAUD implanted radium needles of different strength, filtered through 1 mm. platinum. They were placed in the middle of one testis and removed after various periods. The other testis was removed at the

beginning of the experiment so as to make sure that the animal really was in spermiogenesis. The changes observed in testis after radiation are as follows: Disappearance of spermatogonia within 1 or 2 days, depopulation of the spermatic ducts within 4 weeks with following diminution of testis, and repopulation in 2′ and 3′ month, provided not all the spermatogonia have been killed. — 3 animals that had been treated with 13, 6½, and 3½ Mill. Det. in, respectively, 42, 29, and 30 hours were examined after 52, 41, and 32 days, and showed almost complete sterilization. Still, a few spermatogonia were left. When 15 Mill. Det. was given in 5½ hours, examination 70 days later showed necrosis at the site of application and repopulation in the rest of testis. One animal treated with 4.6 Mill. Det. over 28 days, showed complete sterilization 4 weeks later. From these interesting experiments REGAUD concludes that the radiation time is more important than the size of dosis; protracted irradiations with small doses are more effective than short irradiations with large doses.

It is hardly justified, however, to found a new therapeutic method on these experiments. In the first place, malignant tumors do not necessarily react like testis, even though they have certain points of resemblance; and, secondly, the experiments are not altogether unassailable. Only one testis is treated with a very brief dosis, and only one with a much protracted dosis. The first is examined 70 days after irradiation, the second is examined 28 days after discontinuance of treatment. When it is learned that regeneration sets in just about the 28′ day, the results would have carried more weight if all the animals were examined at one certain time after the discontinuance of irradiation. Finally, in all experiments except the one with very protracted irradiation, the author has removed the other testis in order to make sure that the animal was in spermiogenesis.

However, SCHINZ & SLOTOPOLSKY (156) have made similar experiments with about the same results. They worked with X-radiation on rabbit testes, and they first investigated the effect of different single doses. The results after 100 % and 300 % HED were practically the same. Treatment with 300 % was also followed by regeneration, which began about 4 weeks after treatment. Treatment with 45 % did likewise result in complete depopulation, but was followed by earlier regeneration. Even the effect of 33 % came close to the complete effect of larger doses. In the next experiments the authors gave 100 % and 200 % in single doses and in 3 parts respectively 33.3 % and 66.6 %. The animals were examined at about the same time after the discontinuance of irradiation, and the

result was a more advanced depopulation after the divided doses than after the single ones. As each of the split doses, however, was sufficient by itself to produce depopulation, the authors do not feel justified in concluding that the divided doses were the most effective. They therefore repeated the experiments with 100 % as single dosis and divided it in 5 × 20, 10 × 10, 20 × 5, and 50 × 2 %. The findings were an earlier depopulation and, in particular, a later regeneration after the fractional irradiation. The results appear to be very well-founded.

Later, REGAUD & FERROUX (144) treated rabbit testes with Roentgen rays and found that complete sterilization cannot be obtained with massive doses without giving necrosis of the skin. Splitting the dosis resulted in less effect on the skin and the same, or even stronger effect on testis.

Even before these experiments, the Norwegian investigator MOHR (105) has made a series of excellent experiments, for other purpose, on the testes of Decticus larvæ. Radium radiation was used in 1 × 8, 2 × 5, and 3 × 2 hours. Contrary to others, the author finds that not the spermatogonia, but the youngest spermatocytes are the more radiosensitive cells. Only the latter were affected by 1 × 8 hours; after 2 × 5 hours the older spermatocytes were also affected; and after 3 × 2 hours the spermatogonia, too. This might indicate a stronger action of the divided doses. The objections may be raised to these experiments, which, as mentioned, were made for quite a different purpose, that the number of experimental animals is too small, and that the animals treated with divided doses have lived longer after the beginning of the treatment than the others, what may perhaps have some influence on the result.

There are, besides, a series of very significant experiments on cornea of salamander larvæ by ALBERTI & POLITZER (1, 2, 3). Radiation with 4 H filtered through 0.5 mm. Al produced a so-called primary effect of 6 hours' duration, with rapid fall in number of mitoses and with pycnotic mitoses. Thereafter came a mitosis-free interval of 3 days, and then a secondary stage with increase in mitoses and irregular grouping of the chromosoms. The mitotic picture in the secondary stage was dependent upon the size of dosis; so was the length of the mitosis-free interval. This interval lasted a few hours after treatment with $^1/_3$ H; it lasted 1 day after 1 H, 3 days after 3—4 H, 5 days after 12 H, and 8 days after 20 H. 2 applications of 4 H each gave the following results: If the second dosis was applied 4 hours after the first one, (the primary stage), the effect was the same as after 8 H in one application; if

given after 18 hours (mitosis-free interval), it took longer time fo the mitoses to reappear; if given after 4 days (beginning of secon dary stage), the mitotic figures showed such changes as otherwis are seen after no less than 12 H in one exposure, while the mitosis free interval was still but 3 days; if given after 9 days (height o secondary effect), there was a mitosis-free interval of 3 days, bu the mitotic changes were such as seen after a massive dosis of 20 H From these findings the authors conclude that the better effec is obtained by dividing the dosis in a proper way, and that not only dividing cells are affected but also other cells, whose karyokinesis is delayed, while the mitoses become pathological and mostly lead t necrosis. — Each partial dosis should be just large enough to dest roy the dividing cells, but otherwise as little as possible so as no to prolong the mitosis-free interval. The subsequent doses shoul be given when the secondary effect is at its height.

The difficulty of applying these findings to man is implied i finding the proper time for the second dosis. From theoretical con siderations and from some experiments on the duration of cel division and the various phases of X-ray reaction at various tem peratures, the authors (3) arrive at the conclusion that the prim ary effect in warm-blooded animals presumably is at an end afte 3 hours at the most, and that the mitoses reappear after ca. 1 hours after a large therapeutic dosis.

These findings were confirmed in experiments on mouse em bryos that were irradiated in utero with 9 H through 4 mm. Al. 1 hour after radiation there were numerous mitoses typical of th primary effect, no mitosis after 2 hours, and numerous mitoses — all irregular — after 12 hours. The rational radiation method woul then be 2—3 exposures at intervals of ca. 24 hours, provided tha cancer tissue is affected in the same way as the normal tissues exa mined.

I shall not enter further into the histological changes in cance tissue after irradiation, as that is outside the scope of this work There are numerous papers on that subject, amongst others one b Roussy (147) in Danish. Only the question of mitotic changes afte irradiation will be mentioned in some details.

As early as 1922, before Alberti & Politzer had published their findings, Lacassagne & Monod (87) studied this problem on uterine cancer after radium treatment and on dog sarcoma afte X-radiation with a carcinoma dosis. They found that the mitose were affected at once and disappeared within a very short time After complete rest for some time mitotic figures began to reappear they became more and more frequent, but they were all atypica

and degenerative. In the dog sarcoma the least affected cells were seen to recover their regeneration power; such was not observed in the uterine carcinoma. These excellent studies tally particularly well with ALBERTI & POLITZER's findings.

KREUTER (81), SCHWARTZ (160), HAMPERL & SCHWARTZ (49) have studied this question on Roentgen-treated skin cancers with similar results in the main, although the process ran a somewhat different course with respect to time — with particular reference to the appearance of the secondary stage.

In his investigation on cancer of the uterus after radium treatment DUSTIN (33) has fully confirmed the findings by LACASSAGNE & MONOD.

MOTTRAM, SCOTT & RUSS (109) found similar conditions in Jensen's rat sarcoma after large radium doses. The mitoses disappeared within an hour; on the 4' day the wave of abnormal mitoses was at its height. These mitoses appeared even when the dosis was definitely destructive. One would think, then, that the daughter-cells are incapable of further division.

A particularly interesting series of experiments have recently been reported from The Netherlandish Institute For Cancer Research by STOEL (165). He studied the effect of radium radiation on normal mouse skin, hypertrophic tar-painted skin, tar-papillomas, and tar-carcinomas. In the principle, the mitotic waves were alike in all these cases, but they differed markedly as to duration of the different periods. The time it took before the tissue was free of mitoses was increasing steadily toward the carcinoma state, from 5 minutes in the normal skin to 15 hours in the carcinoma. The mitosis-free interval, on the other hand, was getting shorter and shorter; while the abnormal mitoses appeared in the normal skin after 8—15 days, they were observed after 3—5 days in skin that had been tar-painted 15 times, and after 1—3 days in the carcinoma. The author concludes: »Nur allmählich früher und stärker auftretenden Veränderungen bilden die Unterschiede in der Reaktion zwischen dem Teercarcinom und seine Entwickelungsstadien.»

The same course of mitotic changes after Roentgen radiation have been demonstrated on hen's eggs by REGAUD, LACASSAGNE & JOVIN (145) and in a number of English papers on Roentgen and radium radiation of cultures of chick choroid tissue (STRANGEWAYS & OAKLEY, 167, STRANGEWAYS & HOPWOOD, 166, CANTI & DONALDSON, 22, and CANTI & SPEAR, 23).

The findings by ALBERTI & POLITZER have thus been fully confirmed with regard to the mitotic changes, even though there is some variation in the course of the periods. So there is good reason

to investigate the effect of repeated irradiation in the different periods on malignant tumors.

From the preceding review it is evident that, by various ways and through different materials, the view has been established that split doses, perhaps, still give a better result under certain circumstances than is obtained with massive doses.

B. PREVIOUS INVESTIGATIONS ON TRANSPLANTED TUMORS IN ANIMALS

As introduction, I may mention that Wood & Prime (182) on irradiation of cancer tissue *in vitro* found the lethal dosis to be the same whether it was applied continuously for a short time or it was given in several smaller parts distributed over 12—30 hours. As cancer cells *in vitro* must be regarded as being in a latent state, this result is really quite in keeping with the preceding experiments on plants and animals

There has not been reported very many studies on the effect of split doses on the tumors *in situ*.

Nather & Schinz (123) found inhibition of growth but no cure on massive irradiation of transplanted mouse carcinomas. Treatment with repeated doses at intervals of 2—3 days gave considerable inhibition of growth, and 2 out of 3 animals in one group were cured. From this they conclude that split doses have the better effect. — It may be said to this, that the total dosis in the fractional irradiations has been much larger than the single massive dosis. The two cures, which are the only ones in the whole series, occurred in a group that was treated with 225 % HED × 6, at intervals of 3—4 days, altogether 1,350 % HED unfiltered Roentgen rays within 3 weeks. In massive radiations the highest dosis was 375 % unfiltered and 100 % filtered through 3 mm. Al. The material can not be used for comparison between the effects of equal doses distributed in different ways. Besides, the number of animals (3) in each group is too small to debar accidental factors. — On the other hand, the experiments show that the animals can stand a much larger dose when it is distributed over some time.

Samssonow (154) reports the results of radiation experiments with Roentgen rays on 6 rats with transplanted spindle cell sarcoma from the Radium Institute of Paris. One animal got 5,580 R in a single dosis; the tumor grew more slowly, but the animal died on the 23' day with a large ulceration of the skin. 2 animals got about 9,000 R in smaller portions; the tumors disappeared comple-

tely, but they had not gone at the time when the first animal died. 3 animals got 51—5200 R in 3 applications over 3 days; the tumors disappeared altogether in all the animals, but first after 50—60 days. These experiments, I think, do not warrant the conclusions that the effect is increased by dividing the dosis, as one does not know if the animal that got a large single dosis, might not have recovered, had it lived long enough, and because one single animal is insufficient for control on account of individual variations. — On the other hand, also these experiments suggest that the skin effect is less when the dosis is divided.

In their before-mentioned experiments on Jensen's rat sarcoma, MOTTRAM, SCOTT & RUSS (109) found that 180 millicuries of radium emanation in 30 seconds gave the same mitotic changes as did 30 mgm. of radium salt in 30 minutes, which means a considerably stronger effect from the greater intensity.

Some of the authors reviewed in the preceding chapters have worked with repeated doses, but none of them has compared his findings with the result that may be obtained with a single dosis. So far, then, *there is no accurate experimental basis of estimating the relation between the therapeutical effect of the same dosis on malignant tumors when it is given in one single exposure and when given in split doses.* The following is an attempt to meet this want.

C. OWN INVESTIGATIONS

These experiments are made only on the carcinoma and only after the tumor has come into view. The general technique has been the same as in the preceding experiments. The treatment consisted in local applications to the tumors on femora in the usual manner.

6 S.—N. is used for single dosis, 2 S.—N. × 3 for split dosis. The first series comprises 5 groups, of which the first is treated with a single dosis of 6 S.—N., while the others get 2 S.—N. × 3; the 2′ group is irradiated twice a week, the 3′ group once a week, the 4′ group every 2 weeks, and the 5′ group every 3 weeks.

Treatment of the various groups is started on the same day, and the massive doses are also given on this day. To this arrangement one may raise the objection that the treatment is not completed at the same time in the different groups, so that the experimental conditions apparently are really less favorable to the animals treated at longer intervals, especially when compared with results of single doses, as the fairly rapid growth of tumor implies the

treatment of larger tumors. I have arranged the experiments in this manner because it is the one that most resembles the conditions met with in the clinic. Besides, the split dosis in fractional treatment is always large enough by itself to arrest the growth of tumor for a while, as is evident from my preceding experiments, wherein the average life-time of animals treated with 2 S.—N. is raised from 46.3 to 63.5 days.

The total series are made up of several smaller series, in which the different groups as far as possible contain the same number of equal animals with tumors of the same size. The series then are quite parallel.

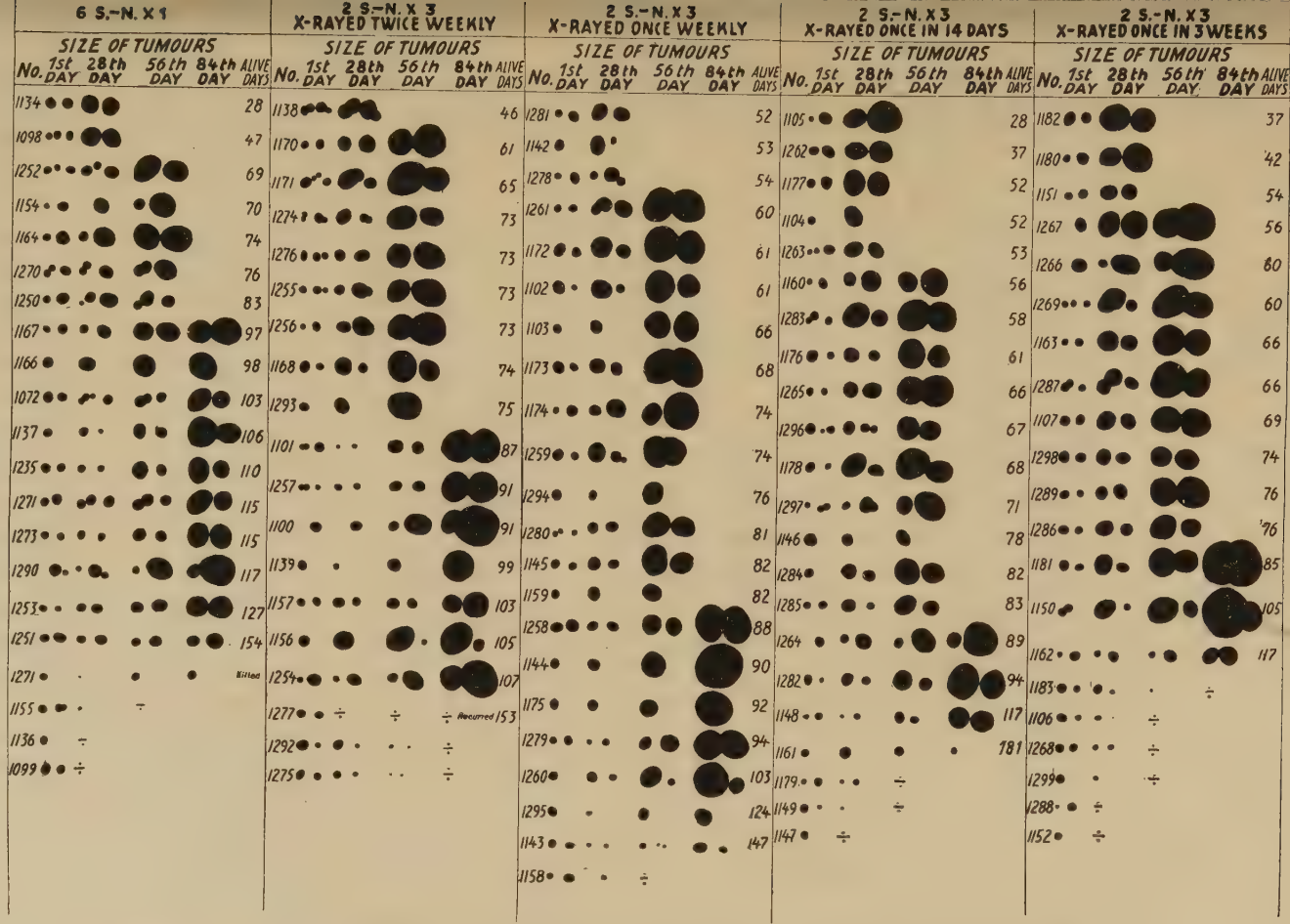
Schema IX shows the outline of the tumors, reduced to 1/4 natural size, at the beginning of the treatment and after 4, 8, and 12 weeks. There is no very marked difference between the various groups. One thing is sure, though, namely that the tumors grow more slowly in the animals treated with a single dosis than in any of the groups irradiated with split doses. Besides, with the possible exception of the group treated every 2 weeks, there seems to be a tendency to increase in rate of growth, the longer the interval between the partial doses.

Table IX gives the numbers of animals in which the tumor has disappeared, and the average life-time of the animals that died with tumors.

Table IX.

	Tumor disappeared in	Average life-time with tumor
6 S.-N. × 1	3 out of 21	93.5 days
		+ 1 animal, killed after 11 months
2 S.-N. × 3, ...	2 » » 19	85.8 days
treatm. given twice weekly		
2 S.-N. × 3, ...	1 » » 22	80.1 »
treatm. given once weekly		
2 S.-N. × 3, ...	3 » » 22	73.3 »
treatm. given once every 2 w.		
2 S.-N. × 3, ...	6 » » 21	69.5 »
treatm. given once every 3 w.		

The number of animals in which the tumor disappeared, is so little that no particular importance can be attached to its variations. Only in one of the groups treated with divided doses is this number larger than that in the group of single doses. This was observed in the last group, where the tumor disappeared in 6 animals that were treated at intervals of 3 weeks. In going through



Schema IX. Carcinoma. Local irradiation with divided doses. Long intervals. Tumors in 1/4 natural size.

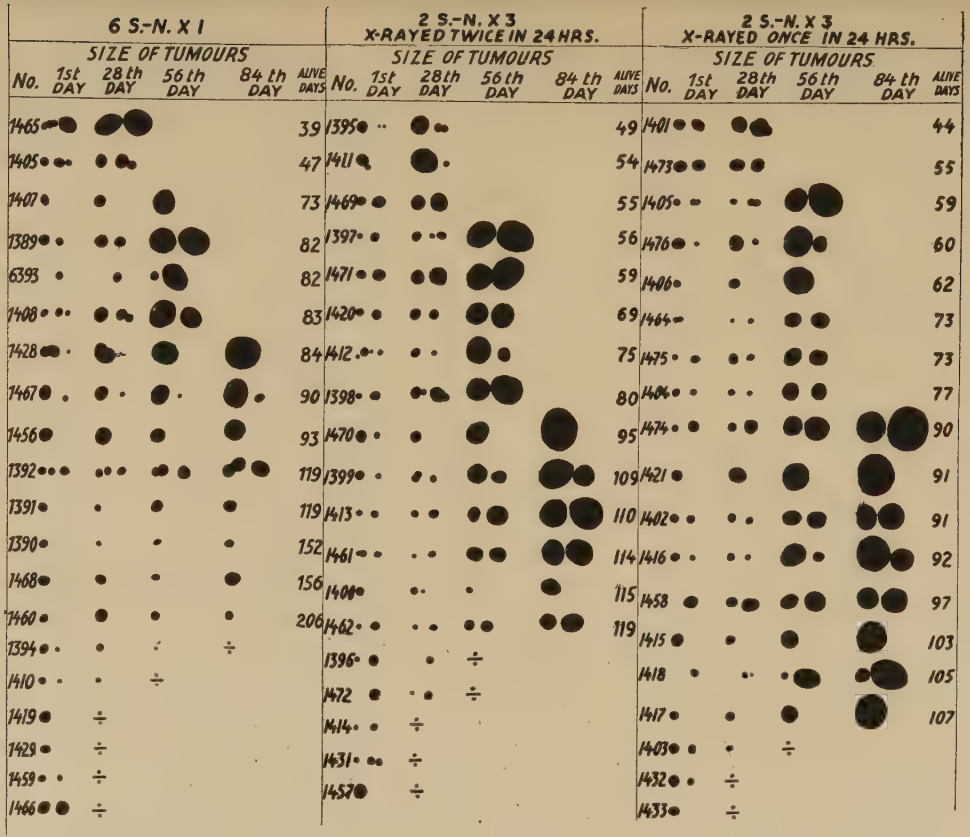
this group, however, it is found that the tumor disappeared in 2 of the animals even before the second treatment, and in the nex 3 before the third treatment; only in 1 animal did the tumor dis appear after the third treatment. For this reason, and because th figures are so small, I do not feel justified in attaching any grea deal of weight to this difference.

The average life-time is found to fall off quite gradually, so th longer the distribution time of the radiation, the shorter the life-tim of the animals. It is particularly noteworthy that the animal which got a single dosis of 6 S.—N. live longer than any of th other groups. This fact is even more pronounced than indicated b the figures: as the average does not include one animal whose tumo (3—4 mm. in diameter) persisted for 11 months without any chang in size. The animal was killed after the 11 months. Microscopi examination showed a tumor of the usual structure, with larg cyst-like dilatations of the alveoli.

Another animal, No. 1277 in group II, should be mentione separately. Its two tumors disappeared soon after the treatment — neither one was palpable in the next 2 months. Then a tumor reap peared on one side, at the very site of the former tumor. It grev rather rapidly and caused the death of the animal in 153 days afte the start of the experiment. Microscopic examination proved it t be a tumor of the same structure as the other tumors, wherefore consider it a recurrence. I have not counted it among the cure animals, but it is entered in the calculations of the average life time.

This experiment comprises altogether 108 animals. The follow ing 3 are omitted from the calculations: 1165, that died 12 day after massive treatment with 6 S.—N.; 1169, that died 7 days afte the experiment had started, and which was to have been treate twice a week; 1153, that was treated once every 3 weeks and die after 77 days with a 3 mm. wide tumor, which on microscopic exam proved to be a carcinoma; but that could hardly have been th cause of death. — Group II has 2 animals less than the othe groups, as I was short of animals in one experiment, so I took onl 2 animals in this group against 4 in each of the other groups.

The animals in which the tumors disappeared, have lived fro then on as follows: Group I, 1 animal $5\frac{1}{2}$ mths., 2 over 6 mths Group II, 1 animal 5, 1 over 6 mths.; Group III, 1 animal $4\frac{1}{2}$ mths Group IV, 2 animals 5 mths., 1 killed after 6 mths. Autopsy on th last one showed no sign whatever of the original tumor, but som pea-sized tumors in each axilla, that on microscopic exam. prove to be carcinomas. I find it more reasonable to take them for pr



Schema X. Carcinoma. Local irradiation with divided doses. Short intervals. Tumors in 1/4 natural size.

mary tumors than for metastases, as I have never seen a simila case of metastasizing, while spontaneous tumors by no means wer exceptional findings among the experimental animals. The tumor were located where spontaneous cancer of the breast often is me with, and the animal was a female. In Group V, finally, 2 anima lived more than 6 months, while the others died respectively 5½ 5, 4, and 3½ months after the disappearance of tumor.

The experiments show that dividing the dosis as mentioned i no instance resulted in a better effect. The effect of the same tota dosis is less when the dosis is divided than when it is given in on exposure; and the effect falls off as the interval between the partia doses is increased.

With a special view to the experiments by ALBERTI & POLITZE I have made an additional series of experiments with shorter inte vals between the partial doses. This makes it fairly probable tha in one of the series, I may have succeeded in hitting the tumor cel while they are in the particularly sensitive state these autho mention.

The same doses are used as in the last experiment, but the pa tial doses of 2 S.—N. are given at intervals of 12 and 24 hour respectively. As in the last experiment, the treatment was initiate at the same time in all the groups. Here, however, the objections this procedure are of less importance, as the tumor can grow b very little in the one day or two the treatment is going on.

The series is made up of several smaller series, in which t animals and the tumors have been alike as far as possible.

Schema X gives the size of the tumors at various times in t usual manner. The rate of growth undoubtedly is less in the grou treated with one single dosis of 6 S.—N. than in the two othe Moreover, the tumors appear to grow a little faster in the grou that is treated once a day than in the group that is treated twice day.

Table X gives the number of animals in which the tumor d appeared and the average life-time of animals that died with tumo

Table X.

	Tumor disappeared in	Average life-time with tumor
6 S.-N. × 1	6 out of 20	101.8 days
2 S.-N. × 3, ... treatm. given twice daily	5 » » 19	82.8 »
2 S.-N. × 3, ... treatm. given once daily	3 » » 19	79.9 »

As shown, the number of vanished tumors is decreasing from Group I to Group III. The life-time shows a similar condition; the difference between Group I and the other groups is very conspicuous.

Thus all 3 criteria of favorable effect are concordant in indicating that, even with these short intervals between the partial doses, the best effect is obtained by giving the total dose at one time.

60 animals were used for this series. 2 animals could not be included in the calculation: 1430, that was treated twice a day; and 1463, that was treated once a day. Unfortunately, they both escaped from their cages.

The life-time of the animals in which the tumor has disappeared was as follows: Group I, 1 animal lived 3 mths., 5 over 6 mths; Group II, 1 animal lived $2^1/_2$ mths., 1 lived 5 mths., 3 over 6 mths.; Group III, 1 lived $5^1/_2$ mths., 2 over 6 mths.

D. DISCUSSION OF THE RESULTS

The results of these experiments are in keeping with the experiments mentioned in the first part of this chapter on growing animal and plant tissue, on unicellular organisms and on human skin, as they show an incomplete cumulation of several repeated radiation doses.

On the other hand, I cannot confirm the view of REGAUD, that protracted treatment with a certain dosis gives a better result than single exposure to the same dosis. Continuous protracted X-radiation being out of the question, I have used divided doses with short intervals, a method that is used in the Roentgen therapy for the same reasons as protracted radiation in the Curie therapy. One might suggest the explanation that the first radiations render the cells resistant to subsequent treatment; and it is possible that the weaker effect from divided doses just is due to an increase of the resistance. But, if so, it will outweigh the possible advantages of protracting the radiation for a longer time. And, if the resistance may be increased within such a short time, there is really no reason, I think, why this should not occur in continuous weak irradiation over the same length of time. As to the theoretical basis of protracted irradiation, that more cells are hit in the mitotic period by this method, it is difficult to estimate the conditions on account of the mitotic changes that have been demonstrated even after small doses. Still, one would think that I have succeeded in hitting more mitotic cells — in some of the groups at any rate — with divided

doses than with the single doses. Nevertheless, I always foun the effect of divided doses to be smaller.

Nor do SCHINZ & SLOTOPOLSKY's findings on rabbit testes seer applicable to the malignant tumors I have used. The same hold good of ALBERTI & POLITZER's findings on salamander larva With the wide variation of intervals in my experiments it is mos likely that I have hit the cells during the period of secondary effec in one of the groups at any rate. It would undoubtedly hav been very interesting to have studied the mitotic details of th tumors I used, but the difficulties of the problem and my ow incompetence have kept me from doing so.

As to the experiments by NATHER & SCHINZ and by SAMSSO NOW, I have already mentioned their shortcomings. If carried ou on a larger material and with a better control, they might hav given some different result. In most instances, besides, these in vestigators gave a larger total dosis with fractional treatment tha on single exposure, a matter I shall later return to.

It might have proved very interesting to look into the cumu lation on irradiation shortly after transplantation, when the cel presumably are in a latent stage. WOOD & PRIME's experiment on cancer tissue *in vitro* showed a complete cumulation of divide doses that were distributed over 12—30 hours. If the same coul be demonstrated on irradiation of the animal shortly after trans plantation, it would mean a strong support of JÜNGLING's theor concerning post-operative treatment of cancer of the breast. Un fortunately, I have not had opportunity of completing these ex periments in the same way as the preceding ones.

These experiments then make against that the mentioned re sults of radiation experiments on testes and salamander larvæ ma be applied to malignant tumors. They indicate, on the other hand *that malignant tumors respond to fractional radiation like most oth growing tissues, as they show an incomplete cumulation.*

These experiments further showed a striking difference in ski effect — estimated by the epilation — between the divided dos and the single doses. Single application of 6 S.—N. would usuall give epilation, while 2 S.—N. × 3 very seldom caused the hai to fall, even when these doses were given at intervals of 12 hour

In the experiments with small intervals epilation occurred i all the animals (19) of Group I, that were treated with a massiv dosis of 6 S.—N. and lived more than 8 weeks after. Only 4 ou of 19 animals in Group II and 2 out of 19 in Group III lost th hairs. In those experiments where epilation occurred in Group

nd III, this effect was always much more pronounced in Group With longer intervals epilation was a very rare exception. — ig. 1 is a photograph of animals from the various groups in the xperiment with the short intervals between the doses.

2 S.—N. × 3, treated once daily | 2 S.—N. × 3, treated twice daily | 6 S.—N. × 1

Fig. 1.

As the relation between effect on tumor and effect on normal issues is the most important question in the therapy, it is there- ore not improbable that protracted treatment — in spite of the emonstrated incomplete cumulation — may yet give a better herapeutic result than is obtained with a massive dosis, as in pro- racted treatment the total dosis may be increased considerably ithout injuring the normal tissues.

The following experiments are meant to throw some light on his question.

VI. ON THE TUMOR EFFECT OF DIFFERENT DOSES WITH THE SAME SKIN EFFECT, GIVEN IN MASSIVE DOSES DIVIDED DOSES, OR AFTER THE SATURATION METHOD

A. PREVIOUS INVESTIGATIONS

As far as I know, there has not before been published any experiment of this kind with comparison in view.

NATHER & SCHINZ (123) and SAMSSONOW (154) verge on the problem, as they think they have obtained better results — with less injury to the surroundings of the tumor — by treatment with several smaller doses. Their experiments have already been discussed in details. REGAUD & FERROUX (144) have treated the scrotum of rabbits with divided doses of Roentgen rays and observed a weaker effect on the skin but the same, or a stronger effect on testes.

As to the saturation method, I have not been able to find any other experiments reported than the undermentioned. KINGERY (71) showed that human skin reacts to irradiation in a way that is in keeping with his theories. PFAHLER (136) thinks he can corroborate the observations made by KINGERY.

WARREN & WHIPPLE (175), on the other hand, conclude from their experiments with irradiation of the abdomen in dogs that KINGERY's formulas do not apply to the mucous membrane of the small intestine. These authors, however, appear to use a ray quality which differs somewhat from the one KINGERY used, and they have only a small number of experimental animals — 5 dogs in all

B. OWN INVESTIGATIONS

First it was necessary to determine the suitable dosage for the different methods so as to get the same effect on the skin.

For criterion of the degree of skin effect I have used the epilation which is easy to observe and really the only dependable sign

f one would go by the erythema, it would be necessary to shave he animal at any rate, and even so it might be rather difficult to listinguish and, in particular, quantitatively to estimate the effect. The ulceration is very variable, I feel, and, besides, one should lso keep in mind that the animals often bite each other.

I have been letting the hairs fall off by themselves, and I have raded the results after the remaining amount of hair: Beginning, partial, and complete epilation.

After medium doses epilation usually comes on in 6—8 weeks, eldom earlier, sometimes later — 10 weeks — and very rarely as ate as 12 weeks after treatment. After larger doses the epilation sually comes earlier and is more complete. After smaller doses it arely is quite complete. After divided doses it usually comes a ittle later than after massive doses.

As epilation may come so late, I have entered only the animals n this calculation that lived more than 10 weeks after the treatment if their hairs did not fall off before.

Like in the preceding experiment, 6 S.—N. is given in single exposure. As a rule this dosis will produce epilation, but in this series of experiments the degree of epilation from this dosis has not been so pronounced as in the preceding experiments. Whether this is because the output of the apparatus has been a little less during this period, or whether the mice have been particularly resistant, that I cannot say. Later the dose showed its old efficacy.

In some preliminary experiments I tried to estimate how long one could give 2 S.—N. twice weekly without producing stronger effect on the skin than is obtained with a single dosis of 6 S.—N. The result was that 2 S.—N. could be given 6 times, altogether 12 S.—N. within 17—18 days.

As to the dosage in treatment after the saturation method I followed PFAHLER's curve for medium-hard rays, wherefore I gave 5 S.—N. for initial dosis and then 2 S.—N. every 5 days, altogether 13 S.—N. in 20 days. The skin effect of this dosage, however, proved to be somewhat stronger than from a single dosis of 6 S.—N., what really was to be expected, inasmuch as PFAHLER's curves for 5 mm. aluminum filter are calculated for a somewhat lower tension than used in these experiments. I decided therefore to extend the intervals but otherwise use the same dosage; and this procedure was found to give a weaker effect.

In the experimental series 5 S.—N. is given at the beginning of the experiment and followed by 2 S.—N. once weekly 4 times, that is a total dosis of 13 S.—N. in 4 weeks. With the large initial dosis one gets close to the tolerance limit at once, and it is aimed to keep

up this effect with the following doses. Smaller intervals are more in keeping with the theory, but I have chosen the longer intervals for practical reasons, so as not to injure the animals too much with the many anesthetics and strappings.

The experiments are made on the carcinoma after the tumor has become palpable. The transplantation is made into femora. The radiations are given locally in the usual manner, and the irradiations of the different groups are started on the same day.

The series is made up of several smaller series with the usual provisions for the number and weight of the animals and for the size of the tumors, so the results are truly comparable.

The following table gives the results of the skin effect, namely the degree and frequency of epilation.

	No epilation	Beginning epilation	Partial epilation
6 S.-N. × 1	16	8	7
2 S.-N. × 6, ... treatm. given twice weekly	24	6	4
5 S.-N. × 1 + 2 S.-N. × 4, ... treatm. given once weekly	15	9	8

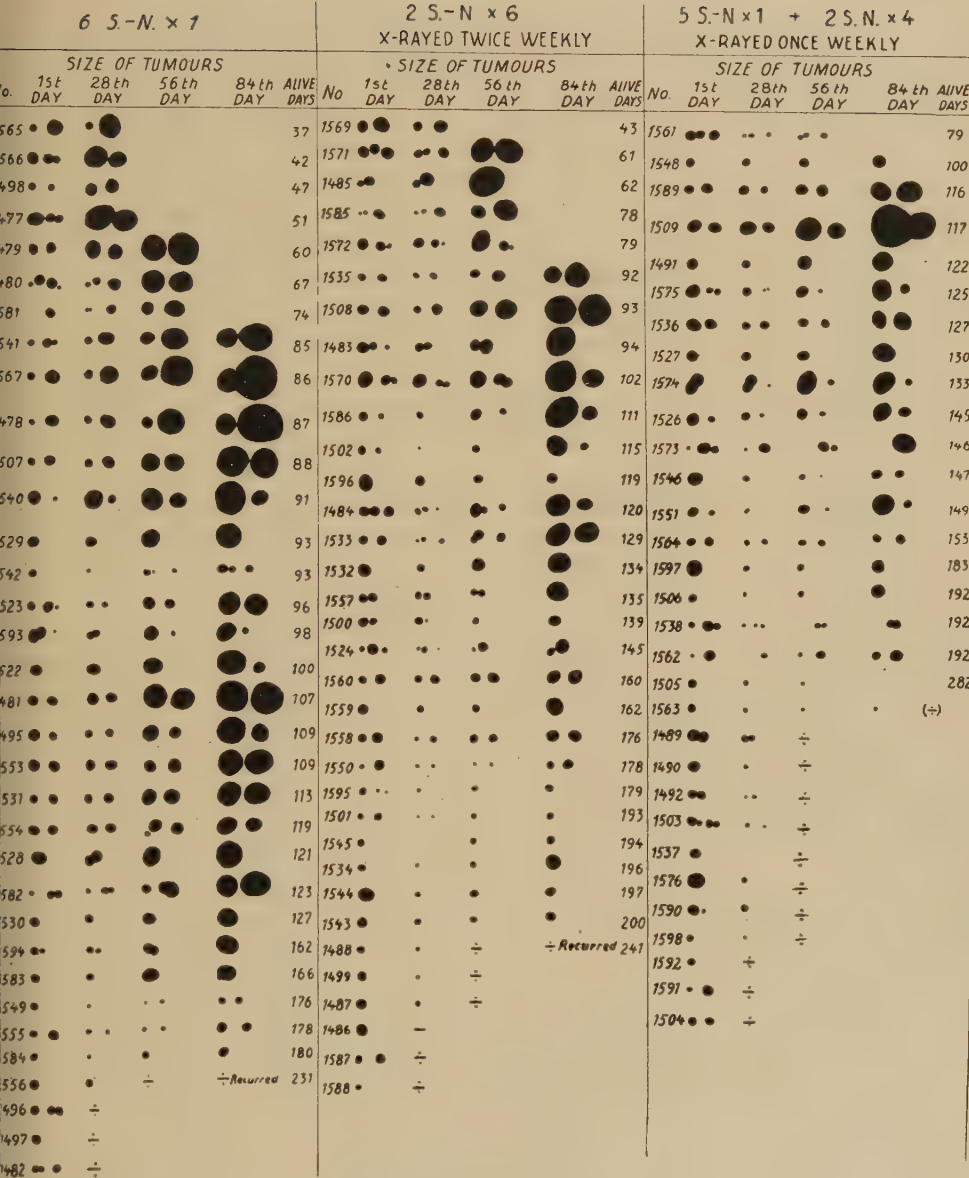
One could hardly expect, I think, to get a more uniform effect than obtained in Groups I and III. In Group II, simple fractional radiation, the effect is less than in the others. One might very well perhaps, give 2 S.—N. × 7 without getting a stronger effect than with a single dosis of 6 S.—N. But, for the subsequent estimation it is important that the fractional treatment does not produce a stronger effect. It was the aim to get a uniform skin effect in the various groups, and that is very nearly accomplished.

Schema XI gives the outlines of the tumors at various times, in the usual manner and reduced to ¼ natural size. There is a very marked difference in rate of growth between Group I and Group II and III in favor of the latter, and there is a distinct difference between Group II and Group III in favor of the saturation method.

Table XI gives the numbers of animals in which the tumor have disappeared and the average life-time of the animals that died with tumors.

Table XI.

	Tumor disappeared in	Average life-time with tumors
6 S.-N. × 1	3 out of 34 — 8.8 %	107.0 days
2 S.-N. × 6	5 » » 34 — 14.7 »	135.4 »
5 S.-N. × 1 + 2 S.-N. × 4 .	12 » » 31 — 38.7 »	148.9 »



Schema XI. Carcinoma. Local irradiation with massive doses, divided doses, and after the saturation method. Tumors in $^{1}/_{4}$ natural size.

The first two groups show no marked difference in the number of animals wherein the tumors have disappeared, merely a suggestion of an increase in favor of the fractional treatment. On the other hand, there is a very marked increase of this number in Group III, where the tumors have disappeared in 12 out of 31 animals.

The average life-time shows a very marked increase from Group I to Group II and an additional, although smaller, increase from Group II to Group III. This increase, however, should be considered on the background of the many surviving animals in Group III. It would, perhaps, be more correct to make the comparison between Group III and the 19 first animals in Group II, which would add very considerably to the difference.

Then all 3 criteria indicate in the first place that, the skin effect being the same, one will obtain a considerably better therapeutic effect with a fractional treatment than with a massive dosis. In the second place they show that the saturation method is superior to the simple fractional treatment.

105 animals altogether were used in this experiment. The following are left out of the account: No. 1568 (in Group I) that died on the 6' day of the experiment; 1539, 1493, 1494 (in Group III) that died with diminishing tumors respectively 7, 19 and 21 days after the experiment was started; 1547 (in Group III) that died after 65 days with a tumor, measuring 3 mm. in diameter, that kept on diminishing till the death of the animal.

After the original tumors in No. 1556 and 1488 had disappeared to palpation, tumors developed anew at the site of the old ones respectively 1½ and 2 months later. These tumors grew rather rapidly and caused the death of the animals respectively in 231 and 241 days after the first radiation. I have taken these tumors for recurrences, as they showed the same microscopical aspects as did the ordinary tumors, and because they developed at the site of the original tumors. So they are included in the calculation of the average life-time.

Of the animals in which the tumors disappeared completely, all but 2 have lived more than 6 months after the tumor had become impalpable. The 2 exceptions belonged to Group III; they died after 2½ and 4 months.

C. DISCUSSION OF THE RESULTS

As to the experiments with simple fractional treatment, the results imply very forcibly that — at any rate in the mouse and

with the tumor employed — the skin recovers its resistance to renewed irradiation more rapidly than the tumor, so that, consequently, one may obtain a better therapeutic effect in proportion to the skin injury with fractional radiation than with single exposure.

Whether the different tumors differ in way of reaction, that is a question to be settled on further investigation. The possibility of such a difference cannot be excluded beforehand.

Another question is whether mouse skin and human skin differ in way of reaction. There can hardly be any principal difference, as all experiments on human skin have shown an incomplete cumulation of repeated doses, just like what is found in all experiments on mouse skin. At most, it will be a matter of quantitative difference.

Beside the skin, other normal tissues — the subcutaneous tissue and the intestinal mucosa, for instance — should of course be taken into consideration. It is by no means settled that these tissues respond to the treatment in the same way as does the skin.

As for the experiments with the saturation method, the skin effect in mice, for one thing, bespeaks that KINGERY and PFAHLER are right in their view of the rate of loss in radiation effect, so that one may give a small dosis shortly after a large dosis and continue in this way for some time without any risk of damaging the skin if only one does not exceed the tolerance limit.

In the next place, the therapeutic results in proportion to skin injury are far better than the results obtained with either of the other methods. This is evident from all the three criteria whereby the results are estimated, and particularly from the number of animals in which the tumors disappeared.

Even the results from a single dosis of 10 S.—N. (p. 37—40), which gives a much stronger skin effect, are no better than the results from the saturation method. Thus it appears that the degree of reaction produced by radiation is not the only important factor in a good therapeutic effect but also the duration of the reaction, so that a weak reaction may be followed by equally good results if only it is kept up for a longer period.

It is PFAHLER's view that the results from the saturation method are better because the reaction is kept up so long that all the cells have passed through the most radiosensitive state. This interpretation is not necessary, however, since a protracted weaker reaction, as mentioned, may very well give a better effect than a stronger reaction of briefer duration, even though the sensitivity of the cells has not changed. That this is so, is obvious from KINGERY's

curves, and it is readily understood through the idea of a biochemical radiation product. If PFAHLER's view were right, one would also expect to obtain a better effect with the same dosis when it is distributed over a longer time. My experiments make strongly against such being the case.

Nor is the assumption of a biochemical radiation product in the tissues essential to the understanding of the value of the saturation method. If we assume that it is the tissue reaction produced by irradiation that acts upon the tumor cells so that the organism for a time has the upper hand in the fight, it is obvious that, if the organism is to win the fight, the reaction must be kept up long enough for it to get all the tumor cells rooted out. This may possibly be accomplished with a very strong reaction for a short while, but it may occur just as well with a weaker reaction over a longer time.

The empirical results, however, stay established, even though the theories be knocked to pieces. The saturation method is a natural and rational combination of the massive dose method and the fractional treatment. To me its theoretical basis, its fundamental train of ideas, appears so agreeable and natural, and I find PFAHLER's results on human beings together with my own results on animals so promising that one is justified in trying the method in many conditions in the clinic, where other methods have given unsatisfactory results.

VII. COMPARISON OF THE EXPERIMENTAL RESULTS AND CLINICAL EXPERIENCES

A tumor in man needs not necessarily react like a mouse tumor; a transplanted tumor is different from a spontaneous tumor; different types of tumors may react differently.

Conclusions from one or two transplanted tumors in white mice to spontaneous tumors in man may therefore be drawn only with the greatest of caution.

In a work of this kind, transplanted mouse tumors offer the advantage of making it possible to obtain large series of uniform tumors. They are preferable to normal tissues of animals or plants, which were often used before in similar studies, as it is still more difficult to draw any valid conclusions from such tissues to malignant tumors in man.

There are many points of resemblance between transplanted mouse tumors and human tumors; and if they are found on some points to react alike to irradiation and other agencies, it is the more likely that they will also react alike on other points.

How do the clinical experiences tally with the findings in experiments on transplanted tumors?

The general macro- and microscopical changes subsequent to irradiation are quite alike, as far as we know.

As to the relation between effect on tumor and size of dosis, my experiments showed an increase in the favorable therapeutic effect when the dosis was increased up to a size that gives rather serious damage of the normal tissues. As far as I know, there has not been reported any reliable observation which proves that this does not apply to man. On the other hand, there are several findings that suggest that in man, too, the best results from local irradiation are obtained with the largest doses, provided that one avoids to injure important neighbouring organs or penetrate too large a part of the organism.

As to the effect of universal irradiation, I found that the doses I have been using, which may be said to be comparatively large,

had no better effect than would be expected from local application of the same dosis. Moreover, it is evident from previous experiments as well as from mine that large universal irradiations may have a pernicious effect. Other investigators, however, appear to have demonstrated that repeated universal irradiations with very small doses may have a good therapeutic effect. — It is reasonable to assume that the effect of the treatment does not rest with the mere universality of the irradiation but that the same result is obtained on irradiation of larger parts of the body.

No clinical report has mentioned a good therapeutic effect from universal irradiation in malignant tumors. Many observations, on the other hand, go to show that irradiation of large parts of the body lowers its resistance to the tumor. This is the most frequent explanation of the poor results in post-operative treatment of cancer of the breast with large massive doses of hard rays applied to the large fields.

The animal experiments with divided doses gave the result that the strongest effect from the same total dose was obtained when the whole dosis was given in one exposure. In proportion to the skin effect, however, a better therapeutic effect could be obtained by giving repeated smaller doses. And the saturation method appeared to be the best way of distributing the doses.

From the present literature it is difficult to judge whether the massive dose method or the protracted treatment offers the best chances of a good clinical result. In this respect, the opinions still differ very much. Yet, there can be no doubt of the trend of progress, as the massive dose method has more and more been replaced by protracted radiation methods, mostly, perhaps, on the score of the general reaction of the patients to the large doses, but often, too, because the results of the protracted method were considered better. It was also taken into consideration that the total dosis could be increased in the protracted method without increasing the effect on the normal tissues.

It is all too early yet to express any opinion of the clinical results from the saturation method. Only this may be said, that PFAHLER's results are promising, and that the words of such an experienced clinician are of considerable weight.

Thus, where the experiences are based on fairly dependable findings, the clinical experiences are really not contradictory to the results from animal experiments, what further justifies the venture of using the experimental results as a guide in the clinic.

If the experiences from this experimental investigation are to be applied to the clinic, the following points may be emphasized

1. The dosis should be as large as practicable without involving any serious injury to important adjacent organs or tissues.

2. Irradiation of large parts of the body with large doses of Roentgen rays should be avoided

3. Distribution of a dosis over a longer time gives less effect than the same dosis distributed over a short time.

4. The fractional or protracted irradiation should be preferred, however, as such treatment allows of a larger total dosis and, therefore gives a better therapeutic effect than may be obtained with the massive dose method.

5. The saturation method is preferable to the simple fractional method.

SUMMARY

1. There is given a brief survey of the radiation methods in irradiation of malignant tumors in man. Special attention has been paid to find out the most adequate size of the dosis, the influence of the general radiation effect on the organism, and the best time distribution of dosis.

On this survey is based the view that the size of dosis has not yet been definitely established, but that it is most probable that the largest doses give the best therapeutic results if the irradiation does not involve large parts of the body, what appears to have an unfavorable influence on the result. No favorable influence from universal irradiation has been demonstrated in the case of malignant tumors.

With regard to the most adequate time distribution of dosis, the various authors disagree very much on the question whether the dosis should be given in the shortest possible time, or it should be distributed over a longer time. In the later years, however, there is a tendency to give more protracted irradiations in contrast to the massive doses which were much in use before. For the present it is not possible from the clinical results to decide which is the best method.

2. After a review of previous investigations with conflicting results into the radiation effect on transplanted tumors in animals, the author reports his own experiments on transplanted tumors in white mice, a carcinoma and a sarcoma. These tumors are treated locally with doses of different size after they have become palpable, or shortly after transplantation.

All the experiments agreed in showing a fairly gradual increase in effect as the dosis was increased, up to twice the size of the dosis that produced epilation in the mice, and which caused a rather severe damage of the normal tissues. This increase in effect was apparent in the rate of growth of the tumors as well as in the average life-time of the animals that died from their tumors, and in the number of animals in which the tumors disappeared or — as in irradiation shortly after transplantation — failed to take.

The discrepancies between previous and own experiments are discussed.

3. Previous experiments on universal irradiation of animals with transplanted tumors are cited. From these experiments it appears that large doses as a rule have an injurious effect, while repeated smaller doses may have a favorable effect. The results, however, are in conflict on this point, too.

The author's own experiments, carried out in the same manner as the preceding but with universal irradiation with various doses, showed no therapeutic effect from the doses employed that came near up to the effect obtained with local irradiation. There is not obtained any stronger effect than could be expected from local application of the same dosis. On irradiation shortly after

transplantation the percentage of takes was increased in proportion to the control. Comparison of the results from universal irradiation with 2 S.—N. and from local treatment with the same dosis showed a better effect from the local treatment. So the universal irradiations have actually done harm in certain cases.

Comparison is made between previous and own experiments.

4. Mention is made of previous investigations on the effects of the same dosis with different time distributions. Some of these investigations are carried out on normal tissues and cells of man, animals and plants, others are done on transplanted animal tumors.

The results do not agree. Most objects with active cell function show a smaller effect when the dose is distributed over longer time; others show the reverse condition. The experiments on transplanted tumors are not conclusive either way.

Own experiments on the carcinoma with local irradiation after development of tumor were consistent in showing a better effect from a single large dosis than from the same dosis distributed in 3 equal parts at intervals of 12 hours to 3 weeks. The skin effect, judged by the degree of epilation, was considerably less after the divided doses than after the same total dosis in one exposure.

The results are discussed.

5. The last series of experiments deals with the therapeutic effects of various doses, that cause the same skin reaction in the mice, and which are given in single exposure, in simple fractional irradiation, and after the saturation method. — There is no previous comparative investigation of this kind.

The experiments are carried out on the carcinoma after development of the tumor. They showed the least effect from the single dosis, greater effect from simple fractional irradiation, and the greatest effect from the saturation method. This was evident from the rate of growth of the tumors, from the number of animals in which the tumors disappeared, and from the average lifetime of the animals that died with tumors.

6. The last chapter gives a comparison between the experimental and the clinical results. The possibilities of using the experiments for a guide in the clinic are discussed, and the conclusive principles for their clinical application are outlined.

RESUMÉ

1. Der er givet en kort Oversigt over Behandlingsmetoderne ved Straale behandling af maligne Tumores i den menneskelige Klinik. Særlig har Opmærk somheden været rettet mod at udfinde den mest hensigtsmæssige Størrelse a Dosis, Indflydelsen af Bestraalingens Almenvirkning paa Organismen og de bedste tidsmæssige Fordeling af Dosis.

Paa Grundlag af denne Oversigt er man kommen til den Opfattelse, a Dosis Størrelse endnu ikke er sikkert fastlagt, men at Sandsynligheden tale for, at de største Doser giver de bedste terapeutiske Resultater, naar man ikk gennemstraaler store Dele af Legemet, hvorved Resultatet synes at paavirke i ugunstig Retning. Nogen gavnlig Indflydelse af universel Bestraaling er ikk paavist for maligne Tumores' Vedkommende.

Med Hensyn til den bedste tidsmæssige Fordeling af Bestraalingerne e der stor Uenighed mellem de forskellige Forfattere om, hvorvidt man skal giv hele Dosis paa saa kort Tid som muligt eller fordele den over længere Tid. De er dog en Tilbøjelighed til i de senere Aar at give mere protraherede Bestraa linger i Modsætning til de tidligere meget anvendte massive Bestraalinger. De er paa nærværende Tidspunkt ikke muligt af Resultaterne i Klinikken at afgøre hvilken Metode der er bedst.

2. Efter en Gennemgang af tidligere Undersøgelser over lokale Bestraa lingers Indflydelse paa transplanterede Svulster hos Dyr, Undersøgelser der ha givet modstridende Resultater, er anført egne Forsøg paa transplantered Svulster hos hvide Mus, et Carcinom og et Sarcom. Disse er bestraalt loka med enkelte Doser af forskellig Størrelse, dels efter at Tumor har udvikl sig, dels kort efter Transplantationen.

Alle Forsøgene viste overensstemmende en ret jævnt tiltagende Virknin efterhaanden som Dosis forøgedes op til en Dosis, der var dobbelt saa st som den, der gav Haaraffald hos Musene, og som gav en ret alvorlig Beska digelse af de normale Væv. Denne tiltagende Virkning viste sig baade i Tu mors Væksthastighed, i Gennemsnitslevetiden af de Dyr, der døde af dere Svulster, og i Antallet af Dyr hvor Tumores forsvandt, eller som ved Bestraalin kort efter Transplantationen ikke fik Anslag.

Uoverensstemmelserne mellem tidligere og egne Forsøg er diskuterede.

3. Tidligere Forsøg over universelle Bestraalingers Indflydelse paa tran planterede Dyretumores er refererede. Det fremgaar af disse, at store Dos som Regel virker skadeligt, medens mindre, gentagne Doser kan have en gavnl Indflydelse. Ogsaa her er dog Resultaterne modstridende.

Egne Forsøg, udført paa samme Maade som de tidligere, men med un verselle Enkeltbestraalinger med forskellige Doser, viste ikke med nogen af anvendte Doser en terapeutisk Virkning, der kom nær paa Højde med d

ved Lokalbestraalinger (med større Doser) opnaaede. Der er ikke opnaaet større Virkning, end man kunde vente af den samme Dosis lokalt. Ved Bestraalingerne kort efter Transplantationen var Anslagsprocenten forhøjet. En Sammenligning af de med 2 S.-N. universelt og de med samme Dosis lokalt bestraalte viste en bedre Virkning hos de udelukkende lokalt bestraalte. Den universelle Bestraaling har saaledes i visse Tilfælde haft en skadelig Indflydelse.

Der er foretaget en Sammenligning mellem tidligere Forsøg og egne.

4. Tidligere Undersøgelser over Virkningen af lige store Doser, fordelt over forskellig lang Tid, er refererede. Det drejer sig dels om Undersøgelser paa normale Væv og Celler af Mennesker, Dyr og Planter, dels om Undersøgelser paa transplanterede Svulster hos Dyr.

Resultaterne stemmer ikke overens. Paa de fleste Objekter i aktiv Cellevirksomhed er der iagttaget en mindre Virkning, naar Dosis fordeltes over længere Tid, men paa andre har man fundet det omvendte Forhold. Forsøgene paa transplanterede Svulster er ikke afgørende i nogen Retning.

Egne Forsøg, foretaget paa Carcinomet med Lokalbestraalinger efter at Tumor er udviklet, viste overensstemmende en bedre Virkning af en enkelt stor Bestraaling end af samme samlede Dosis fordelt i 3 lige store Dele med Mellemrum fra 12 Timer til 3 Uger. Samtidig fandtes en betydelig mindre Virkning paa Huden, bedømt efter Haaraffaldet, af de delte Doser end af Enkeltbestraalingerne.

Resultaterne er diskuterede.

5. I sidste Forsøgsrække er undersøgt Virkningen af forskellige Doser, der gav samme Virkning paa Musenes Hud, og som er givet dels som Enkeltbestraaling, dels som simpel refrakt Bestraaling, dels efter Mætningsmetoden. Tidligere sammenlignende Undersøgelser i denne Retning foreligger ikke.

Forsøgene er foretaget paa Carcinomet, efter at Tumor er udviklet. De viste den mindste Virkning af Enkeltbestraalingerne, større Virkning af simpel refrakt Bestraaling, størst Virkning af Mætningsmetoden. Dette viste sig baade i Svulsternes Væksthastighed, i Antallet af Dyr, hvor Tumores svandt helt, og i Gennemsnitslevetiden af de Dyr der døde af deres Svulster.

6. I sidste Kapitel er foretaget en Sammenligning af de experimentelle Resultater og de kliniske Erfaringer. Mulighederne for Anvendelse af Experimenterne som Rettesnor i Klinikken er diskuterede, og de Principper, man da bør følge i Terapien, er anførte.

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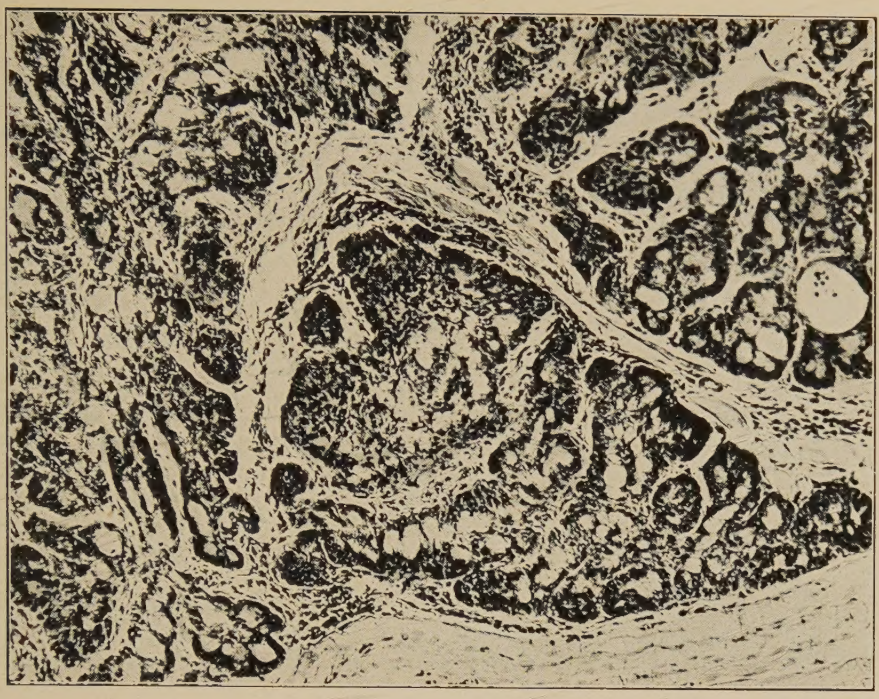
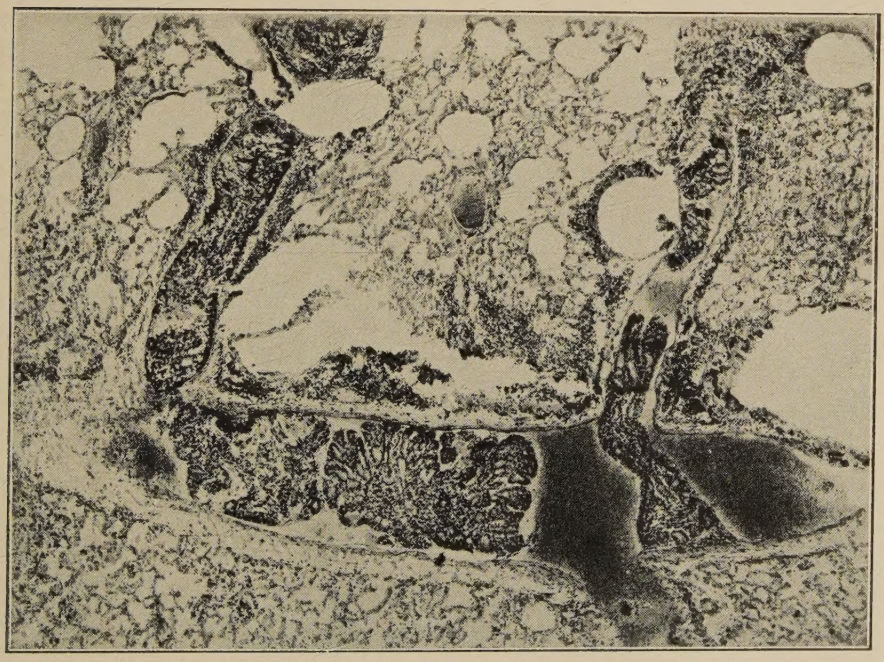
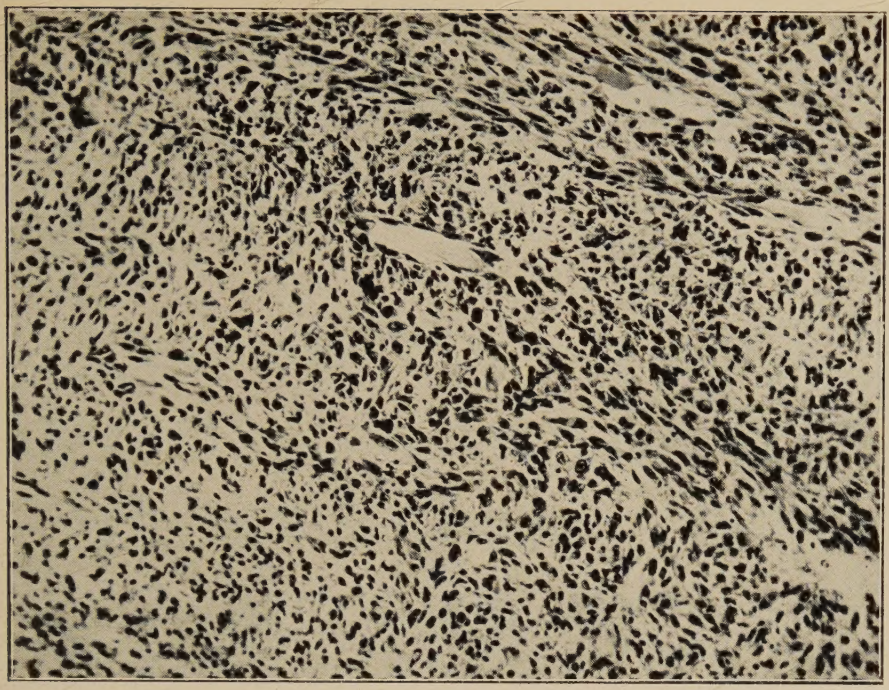
TABULÆ



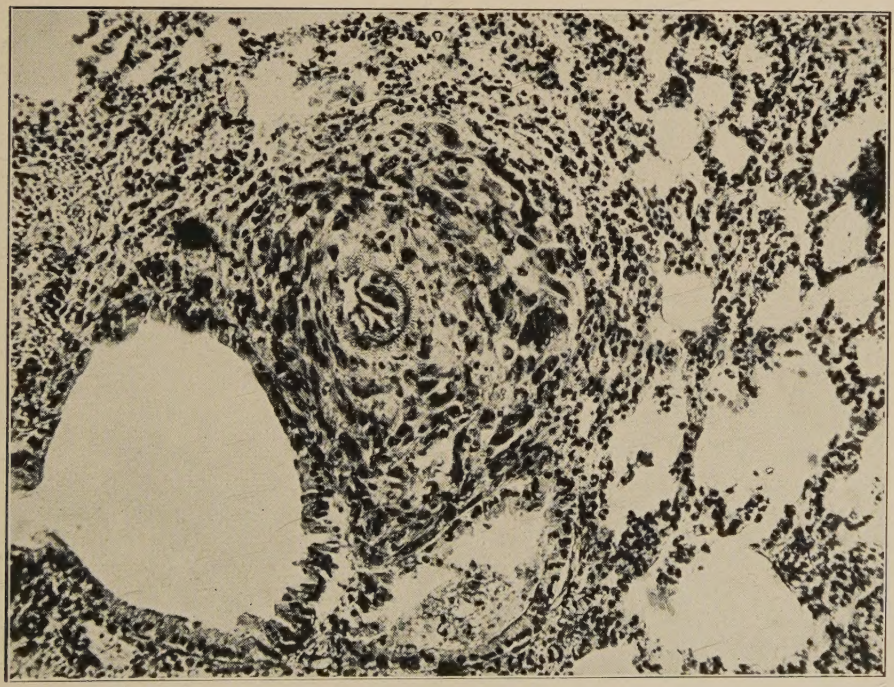
The Carcinoma.



Pulmonary metastasis of the carcinoma.



The Sarcoma.



Pulmonary metastasis of the sarcoma.